

SAMPLING FISH AND INVERTEBRATE RESOURCES IN TIDAL WETLANDS OF THE SACRAMENTO-SAN JOAQUIN DELTA

Report on Phase III Pilot Monitoring: 2017

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September 21, 2018

Executive Summary

The Fish Restoration Program Monitoring Team is tasked with monitoring the effectiveness of tidal wetland restoration in providing habitat and bolstering food web resources for endangered and threatened fishes. The restoration sites are located in the Sacramento-San Joaquin Delta (Delta) and Suisun Marsh (collectively Upper Estuary) pursuant to requirements in the 2008/2009 Biological Opinions for state and federal water project operations. In our initial pilot studies (conducted July 2015 – October 2016 in the north Delta), the primary goal was to determine which methods are reliable and effective for sampling fish and macroinvertebrates in tidal wetlands.

In 2017, we began pre-restoration fish and food web monitoring at future restoration sites throughout the Upper Estuary, as well as in adjacent channels and existing wetlands that serve as reference sites. While conducting the pre-restoration monitoring, we simultaneously addressed questions related to the timing and the amount of effort required for longer-term post-restoration monitoring. The basic questions are:

- 1) Are the fish and pelagic invertebrates collected by existing Interagency Ecological Program (IEP) surveys in deeper parts of the channel representative of the communities in wetland-adjacent shallow water?
- 2) How does the timing of sampling (tidal/diel for zooplankton, different months for macroinvertebrates) affect collections?
- 3) What is the variability in phytoplankton, mesozooplankton, and macroinvertebrates among regions of the Upper Estuary, among site types (tidal, managed, channel), and among habitat types?

Channel and shallow water habitat comparisons

We sampled the fish communities in the shallow water near future restoration sites Prospect Island, Decker Island, and Bradmoor Island/Arnold Slough with either a beach seine or lampara net (shallow water gear types) at the same time the IEP Summer Tow Net (STN; twice per month, June - August) and Fall Midwater Trawl (FMWT; once per month September - December) surveys sampled the closest channel station. Fish communities in similarly shallow water habitats near future restoration site Tule Red were simultaneously sampled with a lampara net and either the STN and FMWT surveys. In addition, fish communities in channel water habitat near future restoration site Winter Island were sampled simultaneously with the lampara net and the STN and FMWT surveys.

During the summer, fish abundance, size, and composition were different between shallow water and channel gear types. The tow net collected smaller fish than the beach seine and lampara net and had lower CPUEs. The beach seine primarily caught American Shad, Mississippi Silverside, Sacramento Sucker, Splittail, Threadfin Shad, and Yellowfin Goby. The most common species caught by the lampara were American Shad, Mississippi Silverside, Striped Bass, Threadfin Shad, and Yellowfin Goby. Tow net catch was dominated by American Shad, Shokihaze Goby, Striped Bass, Threadfin Shad, and *Tridentiger* spp.

Fish catch decreased between summer and fall sampling periods. When comparing the beach seine to the midwater trawl outside Decker Island, CPUE was significantly higher in shallow habitat. Similarly, the lampara CPUE was significantly higher than the midwater trawl when both gears sampled in shallow habitat outside Tule Red. The beach seine's catch was dominated by Mississippi Silverside. The lampara

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net most commonly caught American Shad and Threadfin Shad. The midwater trawl catch was mainly composed of American Shad, Striped Bass, and Threadfin Shad.

Overall, we found differences between sampling gear types, suggesting that habitat and gear type influences the number and fish species caught. Data collected from the channel does not characterize the shallow water habitat fish community. In order to determine what wetland benefits occur after restoration, shallow water sampling by the beach seine and lampara can provide useful fish data such as foraging or rearing patterns not observed by gear types trawling in the channel.

We also sampled mesozooplankton and macrozooplankton in shallow areas adjacent to future restoration sites simultaneously with the channel samples of every other IEP 20 mm survey, which uses a very similar mesozooplankton net and method. Sampling took place once per month March – June. Processing of the 2017 20 mm zooplankton samples is not complete at the time of writing, so the channel – shallow comparison is not included in this report. However, we were able to observe temporal and spatial patterns in zooplankton communities using our own data.

There were major differences in total mesozooplankton catch per unit effort (CPUE) over time and space, with higher catches later in the spring, and higher catches further upstream. Community composition also varied across both time and space. There were higher proportions of calanoid copepods later in the year, and higher proportions of Cladocera further upstream.

There was a small significant effect of distance from the Golden Gate in overall macrozooplankton CPUE, with higher catches further upstream. There was no significant effect of time. Community composition also varied slightly with distance, with more mollusks, annelids, and insects further upstream, but time was not significant. Overall, macrozooplankton were more variable than mesozooplankton, so additional years of data collection may be necessary before conclusions can be reached.

Timing of food web sampling

Our previous work, and other aspects of the 2017 study, focused on *how* and *where* to collect invertebrates that are the primary food sources for our fishes of interest. In order to most efficiently and accurately characterize invertebrate communities, *when* sampling is conducted must also be considered. This study component addressed sample collection on a tidal and diel time scale for mesozooplankton, and on a monthly time scale for mesozooplankton and macroinvertebrates at a single location.

Many zooplankton migrate vertically through the water column, and the movements can correspond to ebb – flood cycles and/or light – dark cues. Over the course of 24 hours and two tidal cycles in June 2017, we examined the temporal variation of zooplankton catches at the water's surface, both over shallow wetland-adjacent habitats and in the center of the channel, and at the spatial variation between the two locations. Additionally, we looked at the differences in catch between channel benthic and surface tows during daylight hours.

We found significantly higher zooplankton abundance at high slack tide, though abundance was higher at night at the other tidal stages. This trend was driven chiefly by the calanoid copepod *Pseudodiaptomus forbesi*, which dominated the zooplankton community. Abundance of zooplankton in benthic tows was slightly higher than surface samples during the day. There were no significant

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differences in community composition or abundance in wetland versus channel habitat in day or night. These results indicate that tidal stage and time of day are important considerations when trying to make inferences on overall wetland productivity from a limited set of zooplankton samples.

Although it may be necessary to sample zooplankton several times throughout the year, larger invertebrates associated with benthic or vegetated habitats are less mobile than zooplankton, and it may only be necessary to sample these macroinvertebrates once or twice per year. If sampling is limited, we want to determine what time of year has greatest overlap between listed fish species and their food supply. We used multiple gears to sample invertebrates at future restoration site Decker Island four times over the course of winter and spring.

We found abundance of macroinvertebrates in all habitat types to increase linearly over the course of the spring. Data from nearby surveys of Delta Smelt and Chinook Salmon indicate that adult smelt are present December through May, peaking in January. Salmon smolts are present in high abundances February through July, peaking in May. Therefore, spring sampling in April should maximize the overlap between macroinvertebrates and the fish that can best make use of these resources. However, this was a single location in a single year, and other areas of the estuary may have different phenologies.

Sample size and variability of food web data

Understanding variability of many food web components will allow us to evaluate appropriate timing and replication of samples, and help us focus monitoring efforts on the most efficient metrics of food web support. To evaluate sample size, we conducted a single, high-replication, spatially intensive sampling effort for zooplankton, benthic macroinvertebrates, epiphytic invertebrates, neuston (surface) invertebrates, chlorophyll-*a*, and phytoplankton at sites distributed across the Delta and Suisun Marsh.

We found that all the ecosystem components we studied were highly variable across space, however some components had greater power than others to differentiate between site types and regions of the estuary. Chlorophyll-*a* concentrations had extremely low within-site variation, but high between-site variation. Future studies should examine temporal variation instead of spatial variation within a wetland. Phytoplankton community composition also varied by region of the estuary and site type, indicating different forms of primary production dominating different areas.

Zooplankton CPUE also had relatively low within-site variation, and high power to detect reasonable differences between sites. We found significant differences in CPUE between regions of the estuary for zooplankton, but not between site types. We found larger differences in community composition both between regions of the estuary and between site types. If 2018 sampling demonstrates similarly high power, zooplankton sample replication may be reduced.

Macroinvertebrates were much more variable than zooplankton, both in CPUE and community composition. While the variability depended on which gear type was being analyzed, even with all habitat types combined the power to detect differences in CPUE between regions was relatively low. Sweep net samples were particularly low-power in detecting differences in CPUE. Unlike zooplankton, macroinvertebrate samples had higher power to detect differences between site types, and we found that wetlands had higher overall CPUE of macroinvertebrates than channel habitat. There were also large differences in community composition between regions of the estuary and site types. However, sampling of some habitats in 2017 was relatively unbalanced, so these data should be combined with data from 2018 before changing sampling replication.

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Preface

Much of the tidal wetland restoration in the Sacramento-San Joaquin Delta and Suisun Marsh (the Upper Estuary) is being constructed under the premise that wetland restoration will increase the resilience of threatened fish. However, drawing empirical connections between restoration and fish is very difficult because of the extreme spatial and temporal variability inherent in estuaries in general, and California estuaries in particular. Given limited funds, we must determine the correct level of replication to answer management questions without cost becoming prohibitive.

The Fish Restoration Program Monitoring Team (FRP) is tasked with developing monitoring plans for tidal wetland sites restored pursuant to requirements in the 2008/2009 Biological Opinions for state and federal water project operations (USFWS 2008, NMFS 2009, CDFW 2009). We led the Interagency Ecological Program (IEP) Tidal Wetlands Monitoring Project Work Team (PWT) in developing the *Tidal wetland monitoring framework for the upper San Francisco Estuary* (hereafter "Framework"; PWT 2017a). The PWT has developed a set of conceptual models and hypotheses for how wetlands benefit fish (Sherman et al. 2017). These were the basis for recommendations for sampling methods to evaluate effectiveness of restoration projects (PWT 2017b). However, there are still outstanding questions as to the appropriate temporal and spatial sampling strategies to test these hypotheses.

Meso- and macroinvertebrates, including amphipods, mysids, insects, copepods, and isopods, are important food resources for tidal wetland fish, but are often patchily distributed and highly variable (Baxter et al. 2015; David et al. 2014; Slater and Baxter 2014; Whitley and Bollens 2014). The spatial and temporal variability inherent in these taxa make them difficult to monitor. While we have already tested several monitoring methods for these groups of invertebrates, monitoring change over time requires understanding the level of spatial and temporal replication necessary for statistical validity. Information on meso- and macroinvertebrates is necessary to address Framework hypotheses F2-F5, which were derived from the PWT's Food Web Conceptual Model (Secondary Production Tier, Hartman et al. 2017a) and Chinook Salmon Tidal Wetland Model (Environmental Drivers and Habitat Attributes tiers, Goertler et al. 2017).

Even for established methods, such as zooplankton trawls, more research is needed to determine the spatial and temporal extent of inference that can be made for a given metric. Multiple long-term monitoring surveys sample the pelagic realm for zooplankton using well-established methods (Hennessy and Enderlein 2013). However, it is unclear the extent to which zooplankton communities differ between the deep channel habitat currently sampled and the wetlands that our program will sample (Bollens et al. 2014; Kimmerer and Slaughter 2016; Kimmerer et al. 2002). Understanding differences in communities between channels and wetlands is also necessary to detect exchange between these habitats that is predicted to increase food availability in the channel (Framework hypotheses F8-F10, Transport Conceptual Model, Hartman et al. 2017b).

Fish are also highly variable across the Upper SF Estuary. Although fishes ranging from larvae to adults are sampled regularly, just a few sampling programs focus on small channel/shallow water/vegetation edge habitats. We have tested several different types of gear for catching fish in these habitats, but thus far have concentrated on a relatively small area of the Delta (Cache Slough). We need a better understanding of the function of these gears across estuarine gradients and how catches compare to long-term IEP monitoring. Fish community sampling in long-term monitoring is needed to determine the

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presence of listed species (hypotheses P4 and P14), to provide specimens of listed fish for various potential studies of diet, condition, and growth (hypotheses F4-F7), and to understand the potential for predation on and competition with the listed species (hypothesis S4), aspects of “capacity” in the Habitat Attributes tier of the Chinook Salmon Tidal Wetlands conceptual model (Goertler et al. 2017).

Pilot Monitoring Phases

We conducted a Phase I “gear exploration” from July to October 2015 (Contreras et al. 2016). Based on results from that effort, successful methods were selected for inclusion in the second phase of pilot work. Phase II occurred from February through July 2016, and provided a more rigorous evaluation of gear feasibility during the time of year listed fish are most likely to be using wetlands (Contreras et al. 2017). Phase II included quantitative comparisons of relative density, diversity, and size distribution of fish caught using the gears selected from Phase I. Methods that were deemed successful from both phases are included in Phase III, described in this report.

After each pilot phase, results were reviewed by the PWT before inclusion in the next phase. The results of all phases will be considered by the PWT in recommending methods for future long-term monitoring. Note that final decisions on the best approaches for long term sampling will be made based on the results of this pilot effort, as well as many additional factors such as take of listed species, logistics, cost, resource availability, and the availability of comparable data from other sampling programs.

Project Objectives

- Determine the extent to which data from IEP’s long-term monitoring surveys can be used to assess fish and invertebrate abundance in shallow, wetland habitat.
- Determine the level of spatial and temporal replication necessary to make sampling design recommendations for long-term monitoring.
- Begin developing a baseline of biomass, community composition, and fish condition for fish and invertebrates near planned tidal restoration and comparison sites. This will allow us to make pre-and-post-restoration comparisons for evaluating restoration progress.

Part 1. Channel versus Shallow Water Comparisons

Introduction

Aquatic science taking place in the Upper SF Estuary can benefit from the wide range of long-term monitoring programs conducted by IEP. The Fish Restoration Program is specifically charged with monitoring tidal wetlands, but nearby long-term monitoring programs' samples may provide adequate data to characterize the ecology of shallow tidal wetlands in the case of some parameters. To characterize variability between channel and shallow-water habitats and reduce likelihood of unnecessary sampling, we conducted a study comparing catch of fish and invertebrates at the edge of wetlands to samples collected in nearby channel habitat.

Shallow water habitat provides benefits to at-risk fish species, such as salmon using it to rear or Delta Smelt inhabiting it to maintain their position during ebb tides when migrating (Bennett and Burau 2015; McLain and Castillo 2009). Shallow water habitat also provides food resources for at-risk fish species. Mesozooplankton, in particular, are a large component of Delta Smelt and salmon diets (Slater and Baxter 2014; Sommer et al. 2001). Macroinvertebrates, including amphipods, cumaceans, insect larvae, and mysid shrimp, are also large components of fish diets (Feyrer et al. 2003; Slater and Baxter 2014). Our conceptual models postulate that tidal wetland restoration sites will have higher production and availability of zooplankton and macroinvertebrates when compared with existing channel habitat and pre-project conditions (Sherman et al. 2017).

However, many of CDFW's long term monitoring studies only sample open water habitat due to gear size, boat size, and absence of vegetation. It is well known that different fish species have different depth and habitat preferences (Young et al. 2018). Furthermore, changes to the physical environment, such as depth and presence of vegetation, will affect the efficiency of our sampling gear. Water depth, substrate, presence of vegetation, presence of benthic grazers (clams), and differences in fish community also alter the zooplankton community composition and abundance (Bollens et al. 2014; Kimmerer and Thompson 2014). Sampling shallow water and open water habitats simultaneously can provide insights into how fish and their food sources utilize different habitats. In this study, FRP sampled fish and zooplankton in shallow water habitat near planned tidal wetland projects concurrently with mid-channel sampling by the IEP 20mm survey, Summer Townet (STN), and Fall Midwater Trawl (FMWT) surveys.

Study Questions:

1. How does the fish community in shallow water habitat compare to open water habitat?
2. How do mesozooplankton communities in wetland and adjacent shallow water habitat compare to open water habitat?
 - a. How do these communities change over the course of the spring?
 - b. How do these communities change along the freshwater to salt water ecocline?

Methods

Fish

Fish were sampled concurrently with the STN and FMWT surveys, June-December, 2017. Sites were chosen based on long-term STN and FMWT sites that are near future tidal wetland restoration locations (Figure 1). Based on the STN and FMWT sampling schedule, sites were surveyed twice a month from June-August and once a month from September-December. Sampling sites were typically void of vegetation and composed of sand and mud substrate mixtures. Sampling site distances between the long-term monitoring surveys to other gear types ranged between 0.3 - 4.3 km.

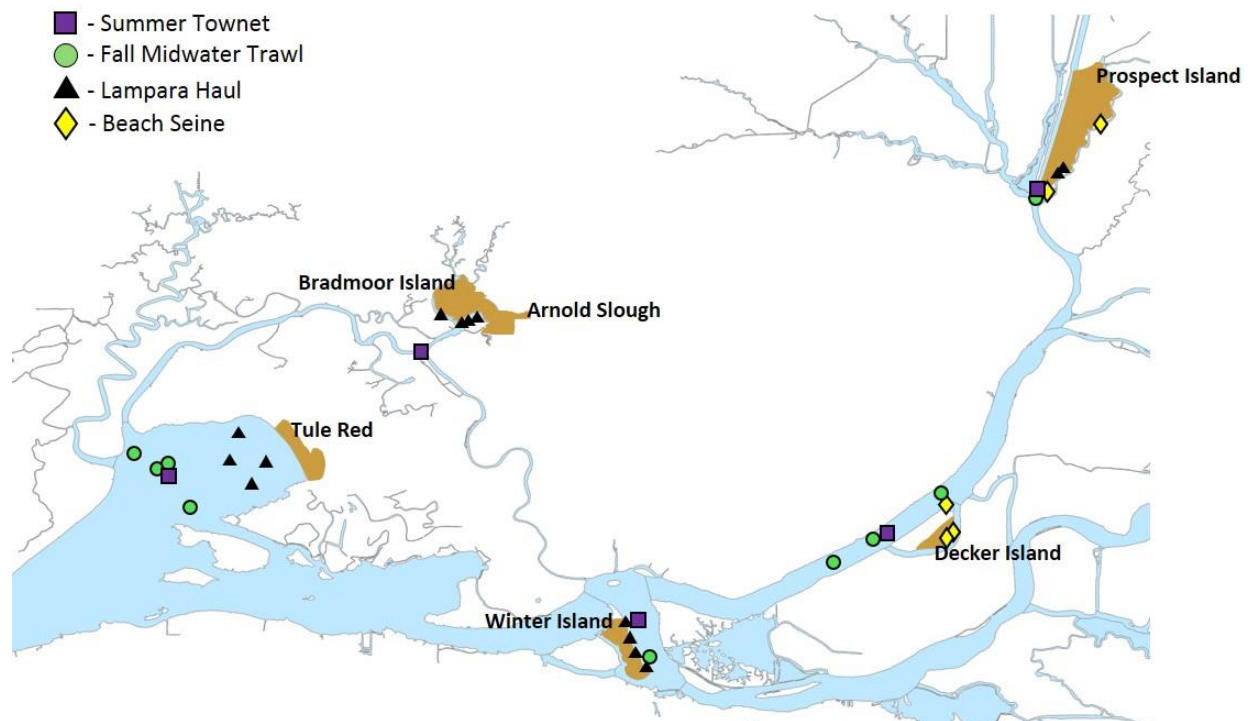


FIGURE 1. GENERAL SAMPLING LOCATIONS. ALL SAMPLING TOOK PLACE NEAR FUTURE TIDAL WETLAND RESTORATION LOCATIONS, INDICATED BY THE LIGHT TAN REGIONS ON THE MAP.

Beach seines and lampara nets were deployed concurrently with the Summer Towntet Survey during the summer (June-August) and with Fall Midwater Trawl during the fall (September-December; Figure 1). We considered concurrent sampling to be when the gears sampled within 4 hours of one another and data were disregarded if they did not fall within this time window.

Before sampling began, the crew scouted all the tidal wetland restoration locations for areas where a beach seine could be deployed. These recurring sites were recorded as waypoints and repeatedly sampled throughout the study. In addition, two lampara sites were fixed in a side channel off Miner Slough because this will be the location of the southern breach of Prospect Island when it is restored as a tidal wetland. All other lampara sampling sites were randomly selected using the sampling design tool in ArcMap (ESRI Inc. Redlands, CA).

Gear Descriptions

Beach Seine: The beach seine is a shallow water gear type that is deployed from shore by crewmembers. It measures 15 m long x 1.2 m high and is composed of 3.2 mm delta square mesh (Figure 2). One crewmember walked perpendicular from shore into the water holding one end of the net until a depth appropriate for proper seining was reached. A second crewmember followed the path of the first crewmember to minimize site disturbance and positioned their seine pole upon reaching the first crewmember. The first crewmember then turned parallel with the shore and continued walking until the seine was fully opened. Water depth and seine lengths were recorded before both crewmembers pulled the seine towards the beach at a similar speed until only the cod end bag remained in the water. The crew filled a tub with water and placed the cod end in the tub along with any fish caught in the wings of the seine. Fork length of thirty individuals of each fish species was measured to the nearest mm and all remaining fish were plus counted. One to three beach seines were completed at Decker Island or Prospect Island outside the tidal wetland restoration area.

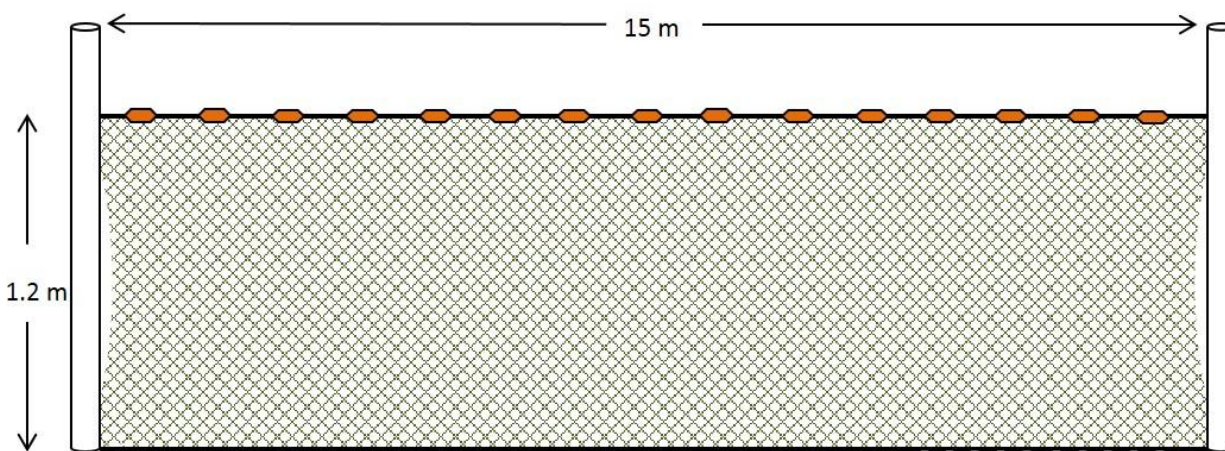


FIGURE 2. BEACH SEINE SPECIFICATIONS.

Lampara Net: The lampara net is a tapered net measuring 36.5 m long x 3.7 m high. The cod end is composed of 9.5 mm stretch mesh and connects to two wings composed of variable stretched mesh (69.9, 146.1, and 88.9 mm, Figure 3). This net was deployed in shallow and channel habitat from a boat where the tip of the wing was tossed into the water attached to a buoy and sea anchor. Crewmembers deployed the net from the bow of the boat as the boat moved in a circular fashion back to the buoy and sea anchor. One crewmember then brought the buoy and sea anchor onboard and hooked both ends of the net onto the front cleat. The boat then went backwards and caused the net to impinge on itself to prevent fish from escaping through the bottom of the net. Once the net was “folded in half”, each crewmember grabbed one side of the net and brought it onboard. Once the cod end was reached, it was placed in a tub filled with water. Fork length of thirty individuals of each fish species was measured to the nearest mm and all remaining fish were plus counted. Three to four lampara samples were completed outside Prospect Island, Winter Island, Bradmoor Island/Arnold Slough, and Tule Red.

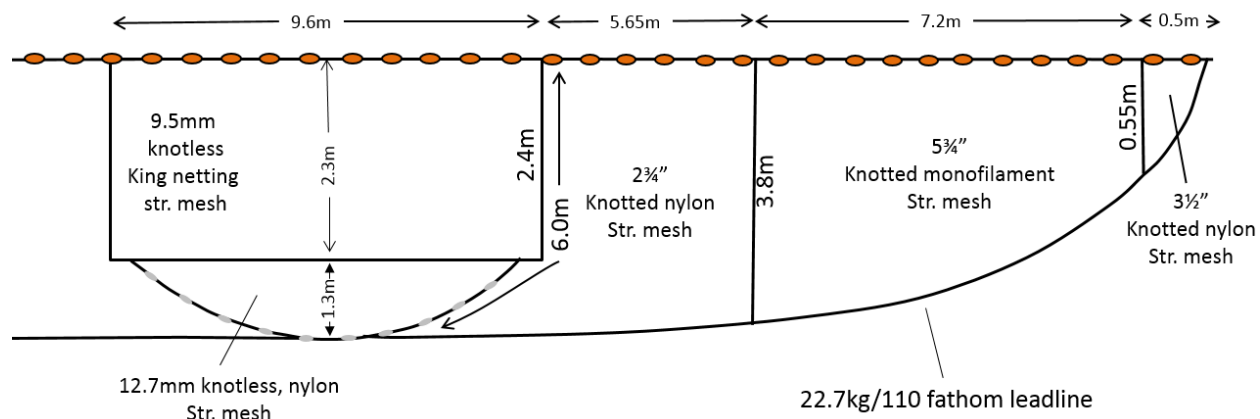


FIGURE 3. LAMPARA NET DIMENSIONS FOR ONE WING AND THE COD END BAG (STR MESH = STRETCHED MESH).

Townet: The townet is composed of 12.7 mm stretch, knotted, nylon mesh 1.8 m long, tapering down to an additional 0.6 m "fyke". This "fyke" fits entirely within the second section, a 2.2 m section of woven mesh with approximately 8 holes per 24.7 mm. The net measures approximately 4.6 m in total, and is lashed directly to a fixed metal "D" frame (Figure 4). The townet was deployed in channel habitat from the stern of a boat. Before each tow, the cod end was tossed into the water when the boat was traveling at an idle speed. The front skis were lifted off the back deck until the net slid off the stern. Once the net and frame had been deployed it free-spooled based on site depth. Once the desired net depth was achieved, hydraulics were engaged and a 10-minute stepped oblique tow began. At the end of the 10-minute tow, the net was brought onboard and fish were released into a tub filled with water. Three tows were conducted if any fish were captured during the first two tows, except in the Sacramento Deep Water Shipping Channel where only two tows were completed. Fork lengths were measured to the nearest millimeter for all Striped Bass and Delta Smelt, and for the first 50 fish per tow for all other species.

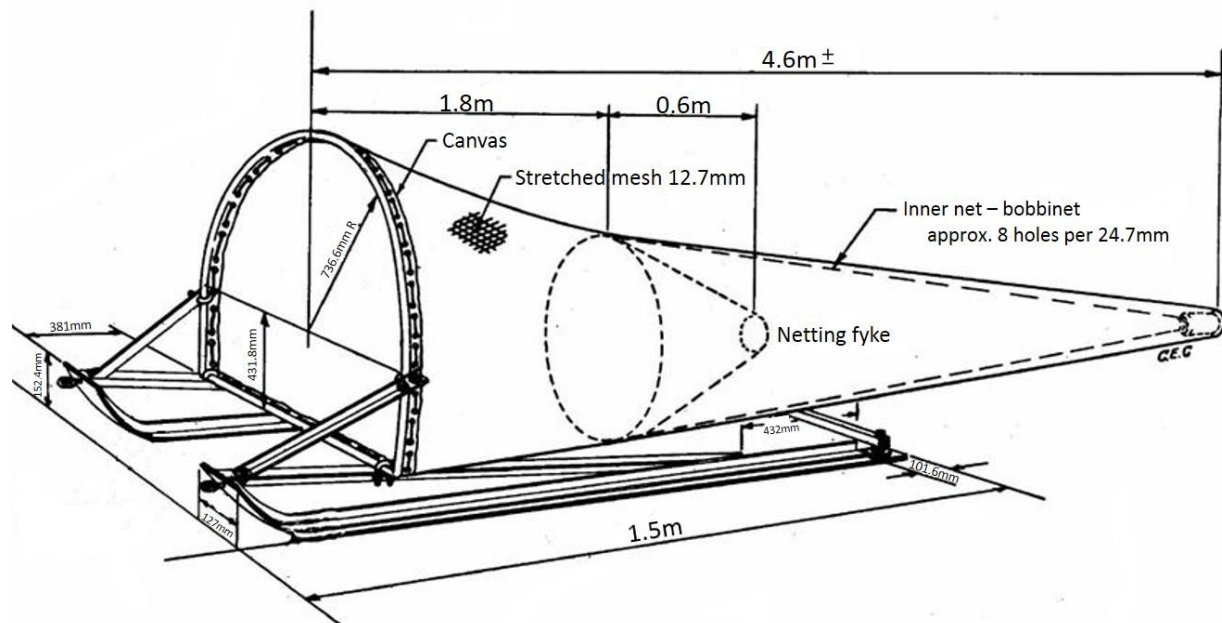


FIGURE 4. TOWNET DIAGRAM SPECIFICATIONS FROM CALHOUN (1953).

Midwater Trawl: The midwater trawl net is approximately 17.7 m long with mouth dimensions of 3.7 m x 3.7 m when stretched taught, but mouth dimensions are smaller when under tension during a tow. Net mesh sizes graduate in nine sections from 203.2 mm at the mouth to 12.7 mm stretch-mesh at the cod-end (Figure 5). The midwater trawl was deployed from the stern of the boat when the boat moved at an idle speed. As the rest of the net was deployed, two crew members each grabbed two planing doors. Each crewmember dropped a pair of doors into the water once the entire net was deployed. The net was then free-spoiled into the water until the site depth was reached, at which a 12-minute continuous tow retrieval began. Once the net was approximately 7.6 m from the stern of the boat, it was brought onboard and all caught fish were released into a tub filled with water. Fork lengths were measured to the nearest millimeter for all Striped Bass and Delta Smelt, and for the first 50 fish per tow for all other species.

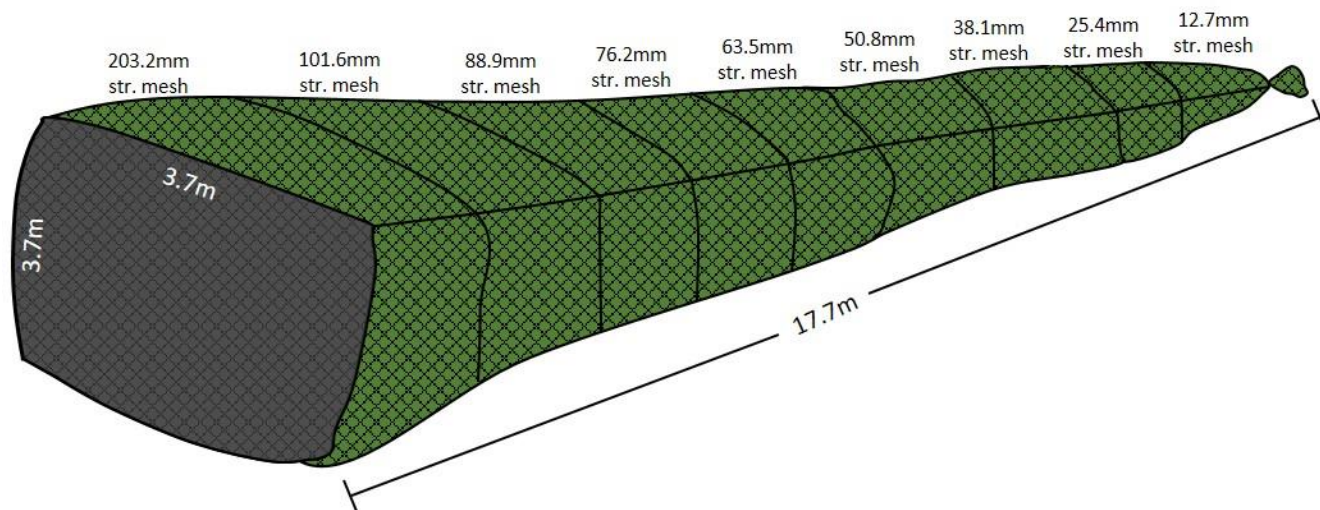


FIGURE 5. MIDWATER TRAWL SPECIFICATIONS DIAGRAM.

Within the context of this paper we refer to the beach seine and lampara net as shallow water gear types and the townet and midwater trawl as channel water gear types. Sampling sites for all gear types outside Tule Red were typically shallow (< 3 m), while sites near Winter Island were typically deeper (> 3 m) (Table 1). Although the depths sampled by the “shallow water gear types” and “channel water gear types” outside Tule Red and Winter Island were similar, the gear types were still compared to look at how gear types may catch/miss various fish species. All other sites provided the desired shallow vs. channel habitat gear type comparisons (Table 1).

TABLE 1. GEAR COMPARISONS MADE AT DIFFERENT SAMPLING LOCATIONS OVER SHALLOW AND CHANNEL WATER HABITAT TYPES.

Months	Sampling Location	Habitat Comparison	Gear Types	n
Jun-Aug	Decker Island	Shallow vs. Channel	Beach Seine vs. Townet	15
Jun-Aug	Prospect Island	Shallow vs. Channel	Beach Seine & Lampara vs. Townet	9
Jun-Aug	Bradmoor Island	Shallow vs. Channel	Lampara vs. Townet	18
Jun-Aug	Tule Red	Shallow vs. Shallow	Lampara vs. Townet	18
Jun-Aug	Winter Island	Channel vs. Channel	Lampara vs. Townet	18
Sep-Dec	Decker Island	Shallow vs. Channel	Beach Seine vs. Midwater Trawl	9
Sep-Dec	Tule Red	Shallow vs. Shallow	Lampara vs. Midwater Trawl	16

Analysis

Three components of data were compared between the sampling gears in each habitat type: fish catch per unit effort (CPUE), fork lengths, and species composition. Fish CPUE was calculated using the number of fish caught per volume water sampled (standardized to 10,000 m³) using the following equation: (fish catch/water volume sampled)*10000. All data were tested for normality using a Wilks-Shapiro test and the appropriate statistical tests were run using Past3 software (Hammer et al. 2001).

Based on results from the Wilks-Shapiro test, a paired t-test or Wilcoxon rank sum test was used to compare CPUEs between the beach seine or lampara to the townet and midwater trawl. Gear

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comparisons were made at each sampling location (e.g., Decker Island, Winter Island, etc). Near Prospect Island, one lampara haul was used to sample shallow water and these CPUE data were combined with beach seine data and compared to channel water gear types' CPUE. Due to a high number of zero catches and low number of samples occurring between the lampara and midwater trawl in channel habitat near Winter Island, no test was run to check for differences between CPUE and fish composition.

A two sample Kolmogorov-Smirnov (K-S) test was used to analyze whether fish size distribution differed between gear types. Three pair-wise comparisons were made independently for the three gear types in the summer and fall. Fish fork lengths were graphed to indicate each gear type's common length ranges and used to set the maximum fork length values to be evaluated. The common size ranges selected represented 94-99% of all fish measured and provided a good representation of each gear's target fish size ranges. Fish sizes larger than the commonly caught size ranges were considered outliers and excluded from analysis. Graphs were made using Excel and R 3.31 with the ggplot2 and plyr packages (R_Core_Team 2018; Wickham 2011; Wickham 2016).

A permutational multivariate analysis of variance (PerMANOVA, (Anderson 2001)) using a Bray-Curtis similarity index was used to test for fish species composition differences between gear types at each sampling location (Table 7, Table 11). Each fish species CPUE was transformed into a percent catch based on the total CPUE for each net deployment. Any fish not identified to species was removed from this analysis except for *Tridentiger spp.* This genus was not removed because identification would have been similar for all gear types. Using the percent catch of each species caught per tow, a PerMANOVA was run using the adonis feature in the vegan package of R (Oksanen et al. 2016) to determine whether differences in fish communities occurred among gear types using the covariates month, temperature, and specific conductance. All samples that caught no fish were removed for the PerMANOVA.

Significance was determined at $\alpha = 0.05$ for all comparisons.

Invertebrates

To address our questions on spatial variation and temporal variation in zooplankton communities, we sampled concurrently with the IEP 20mm survey. The 20mm Survey monitors post-larval and juvenile Delta Smelt distribution throughout their historical spring range in the Sacramento-San Joaquin Delta and San Francisco Estuary. The survey is so named because fish are fully recruited to the gear at lengths >20mm. The survey samples at 40 stations throughout the estuary and completes three 10-minute tows at each station. Zooplankton are also sampled during the first of these tows (Damon, 2015).

The FRP team sampled near seven of these sites in adjacent tidal channels or fringing marsh (Figure 7, Table 12), using paired macrozooplankton and mesozooplankton nets. FRP sampled monthly (every other 20mm survey), in shallow water adjacent to or inside nearby wetlands as close to the same time as possible (usually within 1-2 hours).

Gear Descriptions

20mm Gear: The 20mm Survey net is a cone shaped plankton net 5.1 meters in length with an opening circumference of 4.9 meters (area of 1.5 m²). Zooplankton is collected concurrently with a 160 µm mesh modified Clarke-Bumpus net mounted on the top of the main net frame with its own flowmeter.

FRP Zooplankton Nets: The macrozooplankton net (or “mysid net”) is a 0.4 m x 0.4 m mouth (500 µm mesh size) net that was attached to a steel sled and pulled obliquely through the water column for five minutes. Mesozooplankton were sampled with a 14.6 cm diameter (150 µm mesh size) zooplankton net attached to the macrozooplankton net frame (Figure 6; similar to EMP methods, Hennessy 2009). A flowmeter was mounted in each net to measure sample volume, and effort was standardized by catch per cubic meter of water sampled (see Analysis Methods, below). If the tidal channels or accessible near-shore habitat were too short to take a full five-minute tow, the tow time was reduced. In some cases, gear was held in the mouth of a tidal channel to sample water flowing out of the channel on an ebb tide instead of being trawled.

After retrieval, the nets were rinsed from the outside to wash down the sample into the cod end. All content collected in a cod end was preserved in 70% ethanol for later identification.



FIGURE 6. SET UP OF SLED FOR CONDUCTING OBLIQUE MACROZOOPLANKTON AND MESOZOOPLANKTON TRAWLS.

Laboratory methods

Macroinvertebrate samples: All samples were sorted to extract invertebrates from plant material and detritus, and invertebrates were identified by a Senior Laboratory Assistant (SLA) or Scientific Aide. A subset of samples had identifications checked by an Environmental Scientist for quality assurance. Another subset of samples were checked by an outside lab (Wayne Fields of Hydrobiology), for external quality assurance.

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Subsampling: Approximately 400 invertebrates from each sample were identified. If more than 400 invertebrates were present in a sample, or more than four hours are required for processing, they were quantitatively sub-sampled using a grid tray.

Taxonomic effort: Invertebrates were identified to taxonomic levels corresponding to their importance in fish diets (see Table 2). In the macroinvertebrate samples, mysids, isopods and amphipods were identified to Genus; insects were identified to Family.

Mesozooplankton (Copepoda, Cladocera, and Rotifera) occurring incidentally in the mysid net were enumerated during sorting, but data on these taxa were removed during analysis because they are more accurately quantified using the zooplankton net

TABLE 2. LEVELS OF TAXONOMIC RESOLUTION FOR EACH GROUP OF TAXA COMMONLY FOUND IN INVERTEBRATE SAMPLES. SOME GROUPS WERE IDENTIFIED TO A LOWER LEVEL OF ID IN ZOOPLANKTON (ZOOP) SAMPLES THAN IN MACROINVERTEBRATE (MAC) SAMPLES.

Phylum	Subphylum	Class	Order	Level of ID
Annelida		all	all	Class
Arthropoda	Chelicerata	Arachnida	all	Class
Arthropoda	Crustacea	Maxillopoda: Copepoda	all	Order (MAC)
			Harpacticoida	Order (ZOOP)
			Calanoida	Genus and life stage (ZOOP)
			Cyclopoida	Genus and life stage (ZOOP)
Arthropoda	Crustacea	Malacostraca	Amphipoda	Genus
Arthropoda	Crustacea	Malacostraca	Cumacea	Class
Arthropoda	Crustacea	Malacostraca	Decapoda	Species
Arthropoda	Crustacea	Malacostraca	Isopoda	Genus
Arthropoda	Crustacea	Malacostraca	Mysidacea	Species
Arthropoda	Crustacea	Branchiopoda	Cladocera	Order (MAC)
				Genus (ZOOP)
Arthropoda	Crustacea	Ostracoda	Podocopida	Order
Arthropoda	Hexapoda	Collembola	All	Class
Arthropoda	Hexapoda	Insecta	All	Family
Mollusca		Bivalvia	All	Genus
Mollusca		Gastropoda	All	Family

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Nematoda		All	All	Phylum
Platyhelminthes		All	All	Phylum
Rotifera	All	All	All	Genus (ZOO)

Zooplankton: All samples were filtered and washed in a 150 µm mesh sieve. Filtered zooplankton samples were diluted to a set volume depending on the concentration of zooplankton and/or detritus. 1 mL subsamples were placed on a Sedgewick-Rafter cell glass slide. All organisms were identified to the taxonomic resolution identified in Table 2. At least 5 slides, but no more than 20 slides were processed for each sample, targeting 6% of the total sample. Subsamples were extrapolated to calculate the total number of organisms in the sample in individuals per cubic meter.

All samples were processed by a trained Senior Laboratory Assistant (SLA). A subset of samples had identifications checked by a second SLA for quality assurance.

Analysis

Because the 20mm survey had not completed their spring 2017 sample processing as of June 2018, we analyzed FRP data for temporal and spatial trends, but did not statistically compare to 20mm data. We analyzed mesozooplankton caught in the 150 µm net separately from macrozooplankton caught in the 500 µm net. For each net, we calculated CPUE using the following formula:

$$CPUE = \frac{N}{P * V}$$

Where:

N = Number of organisms counted

P = fraction of sample processed

V = volume of water sampled

And

$$V = (FM_e - FM_s) * k * A$$

Where:

FM_e = Ending flow meter reading

FM_s = Start flow meter reading

k = flow meter constant

A = net mouth area

After examining the CPUE data for outliers, we removed the mesozooplankton sample taken at Lindsey Slough during June, because it contained an extremely high CPUE of rotifers (genus *Branchionus* > 20,000 per cubic meter), that was not seen in any other sample and would cause problems with

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analyses. We assessed the remaining data for differences across the ecocline and over time by performing a generalized linear model on total CPUE for each size class (macrozooplankton and mesozooplankton) using approximate distance from the Golden Gate and month of the year as predictor variables. CPUE was log-transformed to meet assumptions of the linear model (Gotelli and Ellison 2012).

To assess differences in community composition, we performed a PerMANOVA using the “adonis” function in the R package “vegan” (Oksanen et al. 2016) on the percent relative abundance of each major taxonomic group in each sample. Month of the year and distance from the Golden Gate were used as predictor variables. We used Non-metric Multi-Dimensional Scaling (NMDS) to visualize the differences in community composition using the vegan function “metaMDS”, and related the predictor variables to the NMDS using generalized additive models (GAM) with the vegan function “ordisurf” (Oksanen et al. 2016).

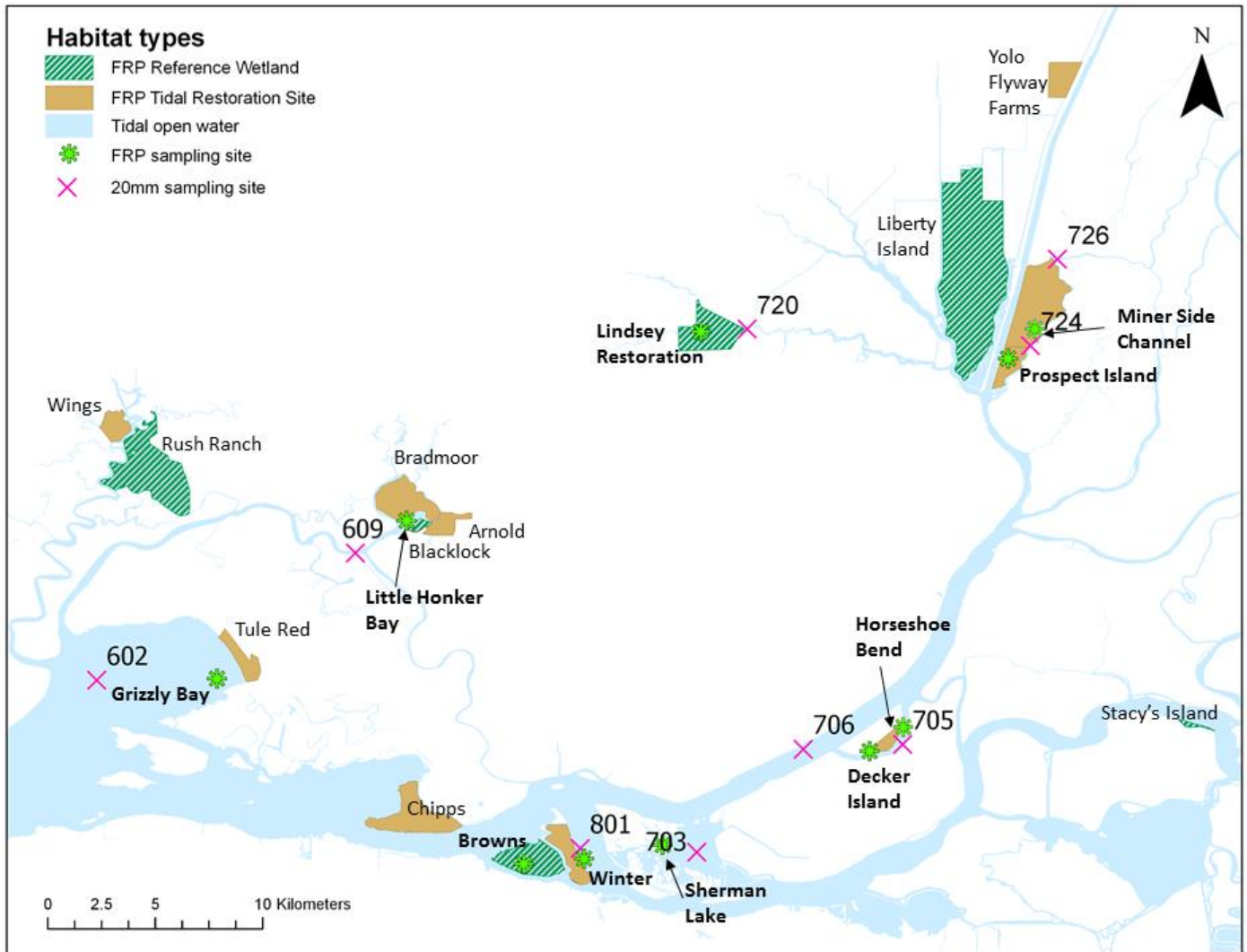


FIGURE 7. ZOOPLANKTON SAMPLING STATIONS. WE TOOK ZOOPLANKTON SAMPLES FROM SHALLOW WETLAND HABITATS (GREEN STARS) IN PROXIMITY TO ESTABLISHED 20MM SAMPLING SITES (PINK XS). FRP RESTORATION AND REFERENCE SITES ARE SHOWN FOR CONTEXT.

TABLE 3. SAMPLE NUMBERS FOR ZOOPLANKTON TRAWLS TAKEN AT ZOOPLANKTON SAMPLING STATIONS. SAMPLES WERE COLLECTED ONCE PER MONTH FROM MARCH-JUNE, 2017, EXCEPT AT BROWNS ISLAND WHERE SAMPLES WERE COLLECTED APRIL-JUNE. THE MESOZOOPLANKTON SAMPLE AT LINDSEY SLOUGH IN JUNE WAS REMOVED FROM THE ANALYSIS DUE TO EXCESSIVE NUMBERS OF ROTIFERS.

20mm Survey Station number	Wetland site	Distance from Golden Gate (km)	Number of Samples	
			mesozoop	macrozoop
720	Lindsey Slough Restoration Site	130	3	4

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724	Prospect Island leaky breach	115	4	4
726	Miner Slough side channel	120	4	4
801	Browns Island	80	4	4
	Winter Island	83	3	3
703	Sherman Island	85	4	4
705	Decker Island	95	4	4
706	Horseshoe Bend	96	4	4
602	Grizzly Bay shoals	65	4	4
609	Little Honker Bay	75	4	4
Total			46	46

Results

Fish

Summer Gear Comparison Between Beach Seine, Lampara, and Townet

A total of 4,592 fish and 25 fish species were collected with fork lengths ranging 7 - 349 mm in 78 pairs of samples (Table 4). When comparing CPUE in shallow vs channel habitats, catch differed between habitat types and was higher in shallow habitat outside Prospect Island, Decker Island, and Bradmoor Island (Table 5; Figure 8A-C). CPUE was also higher in the lampara net compared to the townet in shallow water habitat outside Tule Red (Table 5; Figure 8D). The lampara net and townet had similar CPUEs when both gears sampled in channel water habitat outside Winter Island (Table 5; Figure 8E).

The most abundant fish species caught was the Mississippi Silverside, which made up 51% of the total fish CPUE for all three gear types. Eighty-eight percent of the beach seine's fish CPUE was composed of Mississippi Silverside, Sacramento Sucker, Splittail, Threadfin Shad, and Yellowfin Goby (Figure 11). Splittail and Sacramento Sucker juveniles were primarily collected in June at Decker and Prospect Island by the beach seine. Eighty-seven percent of the total lampara's CPUE consisted of American Shad, Mississippi Silverside, Striped Bass, and Threadfin Shad (Figure 11). Ninety-four percent of the townet's CPUE was composed of American Shad, Shokihaze Goby, Striped Bass, Threadfin Shad, and *Tridentiger spp* (Figure 11).

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TABLE 4. CATCH, CPUE, AND FORK LENGTH (FL) RANGES OF EVERY SPECIES CAUGHT BY EACH GEAR TYPE. A STAR NEXT TO A SPECIES NAME INDICATES NATIVE FISH SPECIES. ALL CAUGHT INVERTEBRATES ARE HIGHLIGHTED AND LISTED AT THE BOTTOM OF TABLE FOR INFORMATIONAL PURPOSES.

Species	Shallow Water Gear Types						Channel Water Gear Type		
	Beach Seine			Lampara			Townet		
	Total Catch	Total CPUE	FL Range (mm)	Total Catch	Total CPUE	FL Range (mm)	Total Catch	Total CPUE	FL Range (mm)
American Shad	53	26939.1	22-41	213	18083.5	16-77	303	3471.0	17-71
Black Bass	15	5444.2	30-67	0	0		0	0	
Black Crappie	1	317.5	110	3	349.7	56-78	2	19.9	45 & 46
Bluegill	3	779.4	27-73	0	0		0	0	
<i>Centrarchid spp.</i>	0	0		0	0		1	11.2	12
Delta Smelt*	0	0		2	311.0	29-39	24	283.2	26-64
Herring UNID	0	0		0	0		12	151.2	25-45
Largemouth Bass	11	2450.0	52-142	1	31.9	265	0	0	
Longfin Smelt*	0	0		1	71.4	55	0	0	
Mississippi Silverside	1217	431386.6	15-89	221	29021.9	22-88	32	366.4	28-67
Mosquitofish	1	210.5	30	0	0		0	0	
Northern Anchovy	0	0		0	0		0	0	19-31
Prickly Sculpin*	16	4643.0	33-90	6	299.2	16-51	0	0	
Rainwater Killifish	83	23649.5	16-38	0	0		0	0	
Red Shiner	2	2040.8	42-51	0	0		0	0	
Sacramento Pikeminnow*	15	7270.4	27-186	2	76.9	172-180	0	0	
Sacramento Sucker*	103	37950.5	17-58	0	0		0	0	
Shimofuri Goby	80	21498.7	18-82	14	2078.7	18-56	13	145.4	15-79
Shokihaze Goby	0	0		1	122.7	33	103	1139.4	15-43
Splittail*	437	99370.3	25-108	16	1461.4	38-297	2	23.7	68-71
Starry Flounder*	0	0		3	451.3	24-55	2	26.5	57-103
Striped Bass	6	684.2	63-90	270	36459.9	14-349	728	8951.9	7-95
Sunfish	3	1554.4	22-25	0	0		0	0	
Threadfin Shad	282	72678.7	19-54	445	51772.0	19-93	160	1848.5	15-96
Three-Spine Stickleback	0	0		0	0		13	128.6	28-37
<i>Tridentiger spp.</i>	0	0		2	201.3	15-17	811	8439.1	10-21
Tule Perch*	3	649.1	54-89	29	1824.0	54-120	1	11.5	64
Unknown	0	0		0	0		77	792.3	7-17
Wakasagi	0	0		4	292.4	52-71	6	83.7	59-81
White Catfish	0	0		0	0		18	267.8	22-77
Yellowfin Goby	92	25441.6	23-95	75	10801.7	20-97	5	62.8	29-83
<i>Crangon spp.</i>	0	0		27	3008.5		22	264.8	
<i>Exopalaemon spp.</i>	284	74681.3		49	5918.4		803	10754.5	
Harris Mud Crab	0	0		19	2851.7		0	0	
<i>Maeotias spp.</i>	0	0		131	13119.1		1563	19133.6	

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<i>Palaemon spp.</i>	0	0		0	0		6	72.0	
Total	2707	839639.7		1534	178608.7		4707	56449.2	
Total (fish only)	2423	764958.5	16-186	1295	153711.1	14-349	1264	26224.3	7-103

TABLE 5. CPUE COMPARISON VALUES FOR GEAR TYPES IN VARIOUS HABITATS.

Shallow vs Channel Habitat Comparisons					
Decker Island (n = 15)					
Gear Type	Mean	Std. Error		Wilcoxon Paired T-Test	
				Z	p
Beach Seine	36094	18777		3.4	0.0007
Townet	285.3	111.2			
Prospect Island (n = 9)					
Gear Type	Mean	Std. Error		Wilcoxon Paired T-Test	
				Z	p
Beach Seine	22762	7931.3		2.5	0.0117
Townet	28.9	21.7			
Bradmoor Island (n = 18)					
Gear Type	Mean	Std. Error		Wilcoxon Paired T-Test	
				Z	p
Lampara	5010.8	1200.3		3.5	0.0003
Townet	818.7	165.2			
Shallow Habitat Comparison					
Tule Red (n = 18)					
Gear Type	Mean	Std. Error		Wilcoxon Paired T-Test	
				Z	p
Lampara	1373	384.5		2.7	0.0073
Townet	306.3	67.9			
Channel Habitat Comparison					
Winter Island (n = 18)					
Gear Type	Mean	Std. Error		Wilcoxon Paired T-Test	
				Z	p
Lampara	294.7	109.2		1.8	0.0684
Townet	84.3	43			

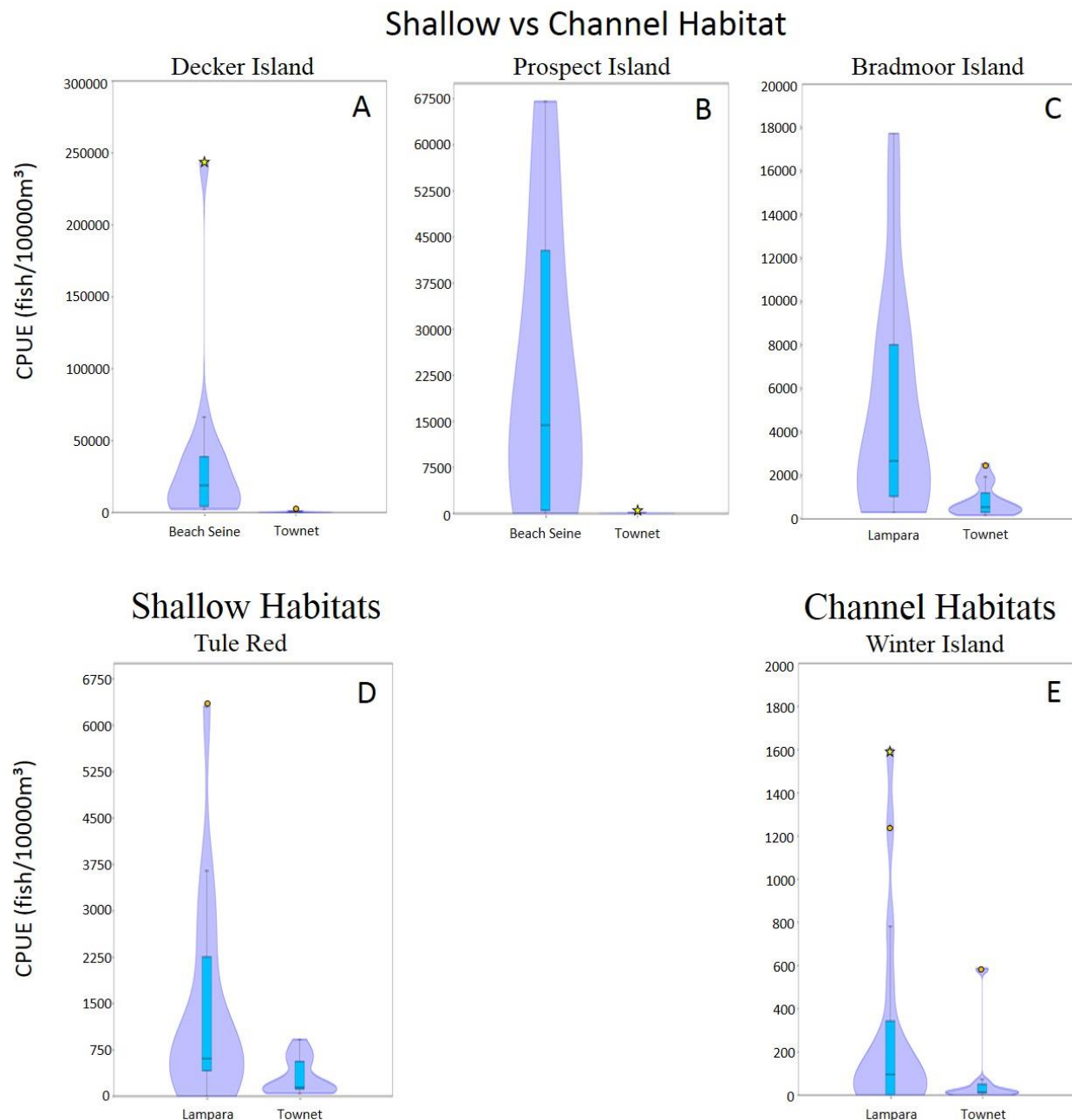


FIGURE 8. CPUE VIOLIN BOXPLOTS OF THE GEAR TYPES IN VARIOUS SAMPLING HABITATS. THE SHADED PURPLE AREAS OVERLAYING THE TRADITIONAL BOX PLOTS INDICATE PROBABILITY DENSITY. THE ORANGE CIRCLES REPRESENT VALUES OUTSIDE THE UPPER INNER FENCE AND YELLOW STARS REPRESENT VALUES 3 TIMES THE BOX HEIGHT FROM THE BOX.

All three Kolmogorov-Smirnov tests showed significant differences of fish lengths between all three gear types (Table 6), however the towner consistently caught smaller fish primarily comprised of *Tridentiger spp.* and Striped Bass (Figure 9). Within the context of this comparison, the beach seine captured a wide range of juvenile Shimofuri Gobies, Splittail, and Sacramento Suckers; the lampara net caught a wide range of lengths for American Shad, Mississippi Silversides, Striped Bass, Threadfin Shad, and Yellowfin Goby; the towner caught a wide range of lengths for American Shad, Striped Bass, and Threadfin Shad (Figure 10).

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TABLE 6. KOLMOGOROV-SMIRNOV COMPARISONS OF FISH SIZES BETWEEN GEAR TYPES.

Beach Seine		Lampara	
N:	935	N:	875
D :	0.15347	p:	8.81E-10
Beach Seine		Townet	
N:	935	N:	1814
D :	0.45654	p:	3.04E-113
Lampara		Townet	
N:	875	N:	1814
D :	0.45932	p:	1.05E-109

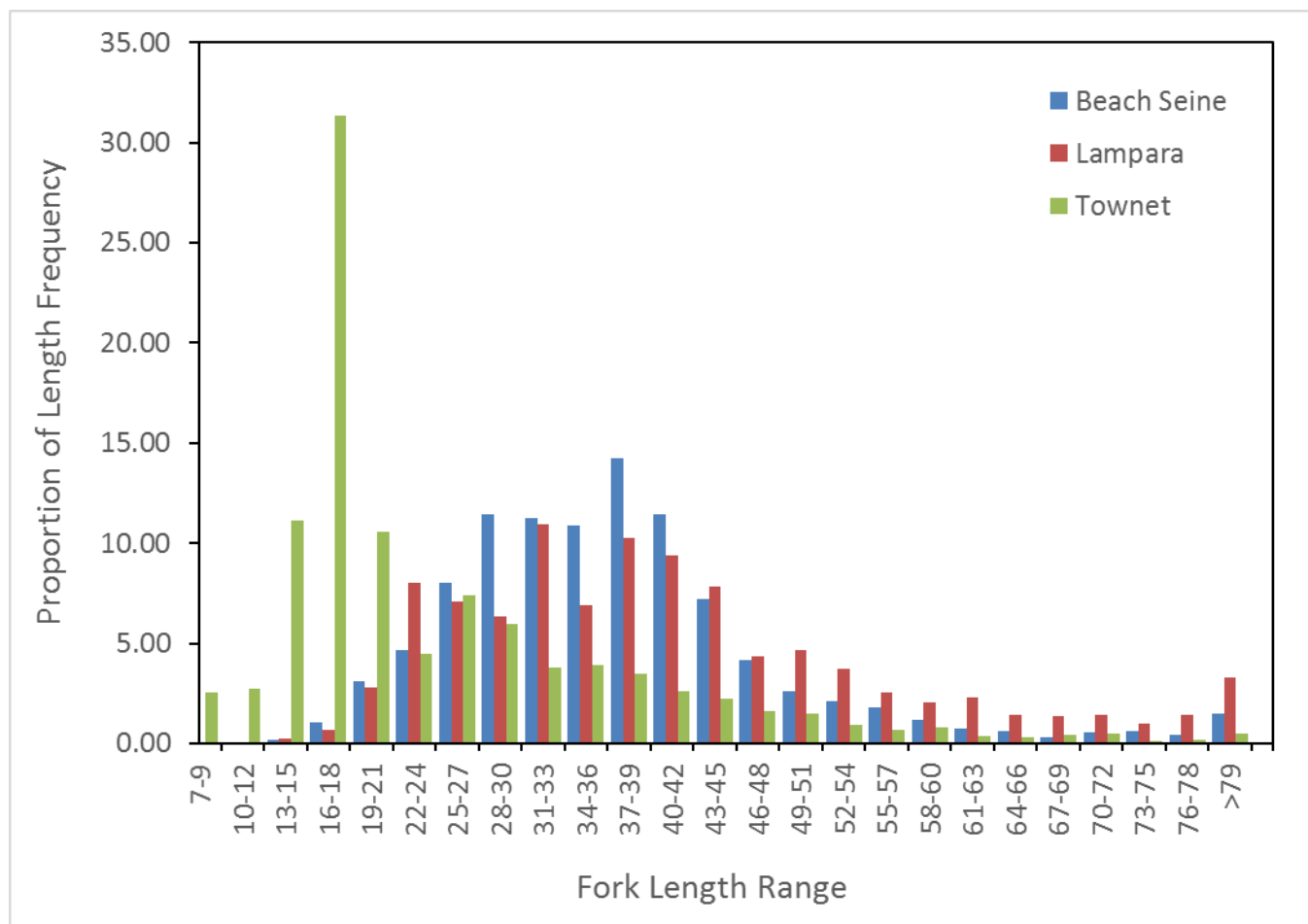


FIGURE 9. FORK LENGTHS CAUGHT BY EACH GEAR TYPE. FISH GREATER THAN 78 MM WERE NOT USED FOR LENGTH COMPARISONS BETWEEN GEAR TYPES, BUT ARE SHOWN HERE FOR ADDITIONAL INFORMATION.

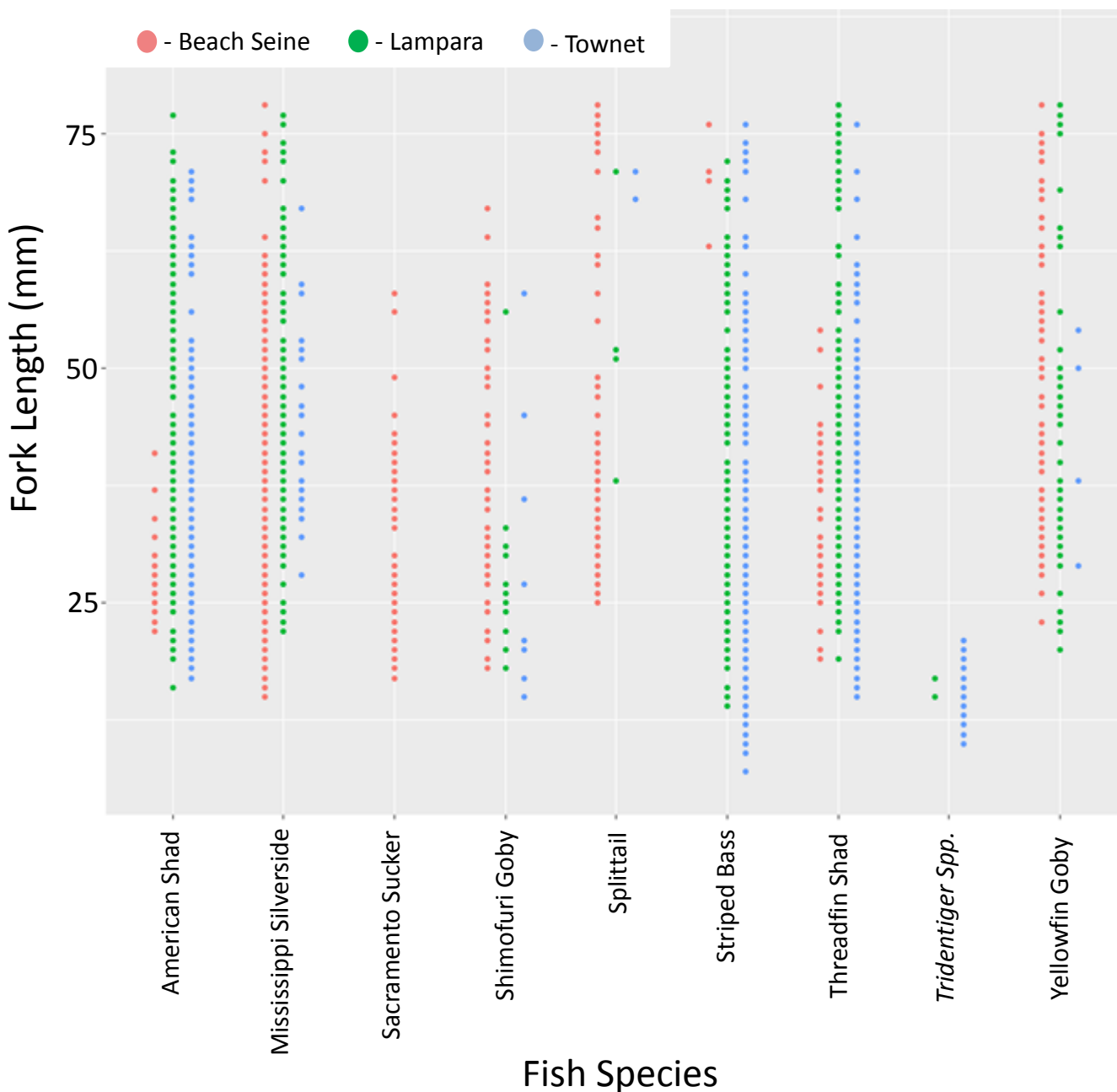


FIGURE 10. FORK LENGTHS FOR COMMON FISH CAPTURED IN THE BEACH SEINE, LAMPARA, AND TOWNET. EACH DOT REPRESENTS AN INDIVIDUAL FISH LENGTH CAUGHT BY EACH GEAR TYPE.

Each gear type caught between 15-17 fish species (Table 4) and all habitat comparisons resulted in differences in fish composition due to gear type (Table 7). However, the time of catch (i.e., covariate – “month”) was also a predictor of fish composition differences between gear types at Bradmoor Island and Tule Red (Table 7). In general, the townet caught a higher abundance of Striped Bass, Tridentiger gobies, and White Catfish (Figure 11). The beach seine caught a higher abundance of Mississippi Silversides and Splittail (Figure 11). The lampara net generally caught higher abundances of Mississippi Silverside, Yellowfin Goby, American Shad, and Threadfin Shad (Figure 11). The townet also caught a higher number of Delta Smelt when sampling shallow water habitat outside Tule Red.

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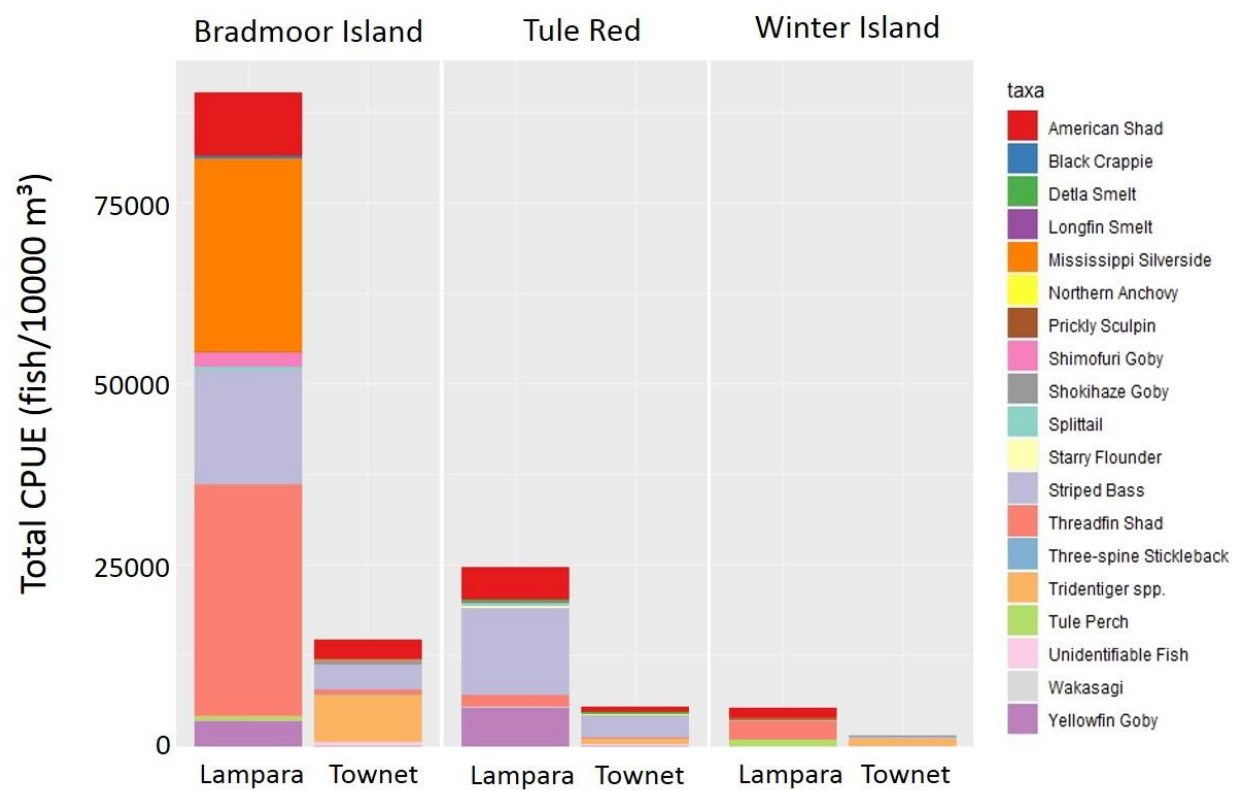
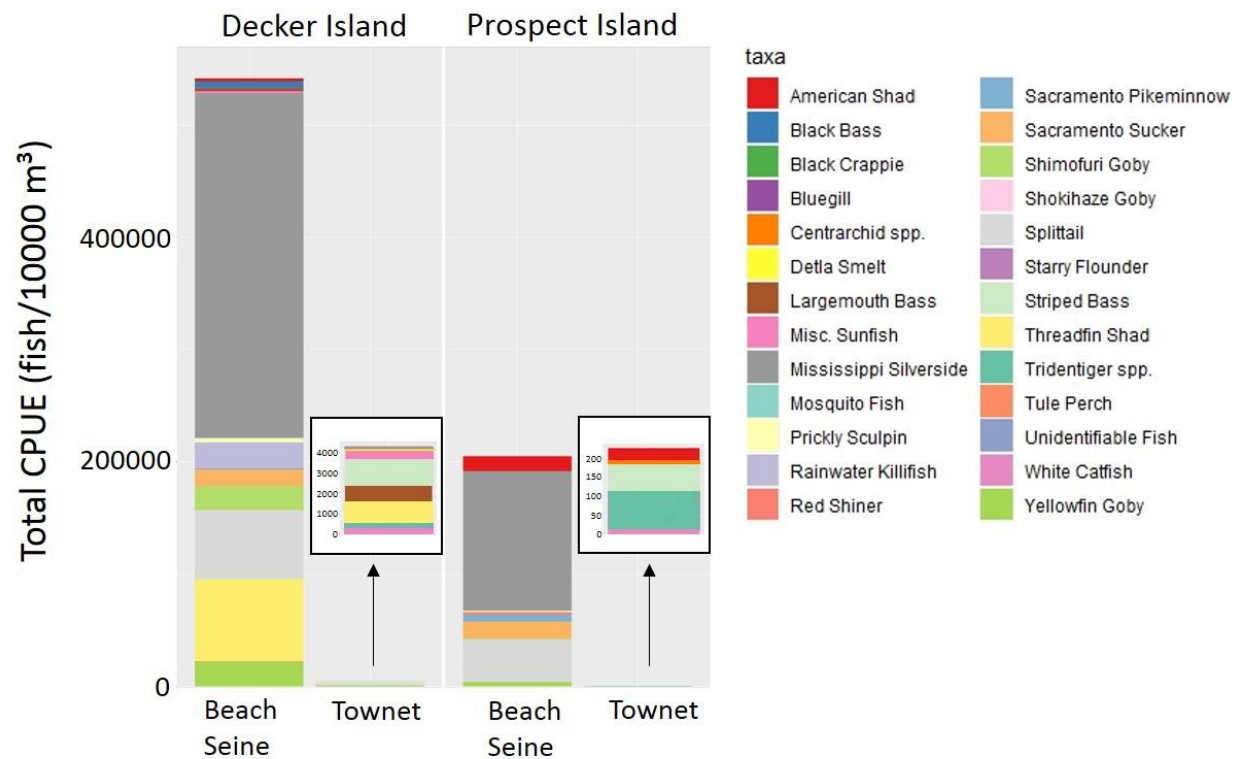
TABLE 7. RESULTS OF PERMANOVAS ON RELATIVE ABUNDANCE OF FISH SPECIES DURING THE SUMMER TOWNET SURVEY. ALL HABITAT COMPARISONS SHOW DIFFERENCES OF FISH COMPOSITION INFLUENCED BY GEAR TYPE AT EACH SAMPLING LOCATION.

PerMANOVA Channel vs Shallow Water - Decker Island						
	Df	Sums of Sqs	Mean Sqs	F-value	R ²	p-value
Gear.Type	1	3.5833	3.5833	20.339	0.37503	0.01 **
Month	1	0.7841	0.7841	4.4507	0.08207	0.01 **
Temp	1	0.6159	0.6159	3.496	0.06446	0.01 **
SpC	1	0.343	0.343	1.947	0.0359	0.11
Residuals	24	4.2282	0.1762		0.44254	
Total	28	9.5545			1	
PerMANOVA Channel vs Shallow Water - Prospect Island						
	Df	Sums of Sqs	Mean Sqs	F-value	R ²	p-value
Gear.Type	1	0.9665	0.96655	2.41027	0.17743	0.02 *
Month	1	0.477	0.47704	1.18959	0.08757	0.19
Temp	1	0.4384	0.43837	1.09315	0.08047	0.38
SpC	1	0.3573	0.35732	0.89104	0.06559	0.63
Residuals	8	3.2081	0.40101		0.58893	
Total	12	5.4474			1	
PerMANOVA Channel vs Shallow Water - Bradmoor Island						
	Df	Sums of Sqs	Mean Sqs	F-value	R ²	p-value
Gear.Type	1	1.6887	1.68867	12.2358	0.18815	0.01 **
Month	1	2.4083	2.40832	17.4502	0.26834	0.01 **
Temp	1	0.322	0.32199	2.3331	0.03588	0.1
SpC	1	0.2777	0.27771	2.0122	0.03094	0.15
Residuals	31	4.2783	0.13801		0.47669	
Total	35	8.975			1	
PerMANOVA Shallow Vs Shallow Water - Tule Red						
	Df	Sums of Sqs	Mean Sqs	F-value	R ²	p-value
Gear.Type	1	0.6145	0.61446	4.6474	0.08017	0.01 **
Month	1	2.825	2.82501	21.3662	0.3686	0.01 **
Temp	1	0.2919	0.29194	2.208	0.03809	0.08
SpC	1	0.0985	0.09851	0.7451	0.01285	0.47
Residuals	29	3.8343	0.13222		0.50029	
Total	33	7.6643			1	
PerMANOVA Channel Vs Channel Water - Winter Island						
	Df	Sums of Sqs	Mean Sqs	F-value	R ²	p-value
Gear.Type	1	3.4941	3.4941	15.6992	0.34643	0.01 **
Month	1	0.4578	0.4578	2.0572	0.04539	0.1
Temp	1	0.4502	0.4502	2.0227	0.04463	0.14

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SpC	1	0.3425	0.3425	1.5387	0.03395	0.21
Residuals	24	5.3415	0.2226		0.52959	
Total	28	10.0861			1	

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FIGURE 11. STACKED BAR CHART OF THE TOTAL CPUE OF FISH SPECIES CAUGHT BY EACH GEAR TYPE OUTSIDE FUTURE RESTORED TIDAL WETLANDS DURING SUMMER. AT DECKER AND PROSPECT ISLANDS, AN ENLARGED STACKED BAR GRAPH APPEARS ABOVE THE TOWNET STACKED BAR CHART TO SHOW WHICH FISH WERE CAUGHT.

Fall Gear Comparison Between the Beach Seine, Lampara, and Midwater Trawl

Fish catch decreased between summer and fall sampling periods. A total of 2,863 fish and 21 fish species were collected with fork lengths ranging from 20-454 mm from 25 pairs of sites (Table 8). When comparing the beach seine to the midwater trawl outside Decker Island, CPUE was significantly higher in shallow habitat (Table 9; Figure 12A). Similarly, the lampara CPUE was significantly higher than the midwater trawl when both gears sampled in shallow habitat outside Tule Red (Table 9, Figure 12B).

Ninety-eight percent of the beach seine's total fish CPUE was composed of Mississippi Silverside. Eighty-six percent of the lampara's total fish CPUE was composed of American Shad, Mississippi Silverside, and Threadfin Shad. Ninety-four percent of the midwater trawl's total fish CPUE was composed of American Shad, Striped Bass, and Threadfin Shad. All gear types caught invertebrates (Table 8), but this data was not used for analysis.

TABLE 8. CATCH, CPUE, AND FORK LENGTH (FL) RANGES OF EVERY SPECIES CAUGHT BY EACH GEAR TYPE. A STAR INDICATES IT'S A NATIVE SPECIES. ALL CAUGHT INVERTEBRATES ARE HIGHLIGHTED AND PLACED AT THE BOTTOM OF TABLE FOR INFORMATIONAL PURPOSES.

Species	Beach Seine			Lampara			Midwater Trawl		
	Total Catch	Total CPUE	FL Range (mm)	Total Catch	Total CPUE	FL Range (mm)	Total Catch	Total CPUE	FL Range (mm)
American Shad				45	2562.6	58-91	374	644.6	42-109
Delta Smelt*				3	190.8	48-55			
Jacksmelt*							1	1.8	257
Largemouth Bass	3	628.9	70-225						
Longfin Smelt*							13	23.9	52-86
Mississippi Silverside	1786	319851.4	24-95	11	631.3	49-87			
Mosquitofish	2	227.8	22-27						
Northern Anchovy*							13	21.9	60-84
Rainwater Killifish	9	1499.4	20-30						
Sac. Pikeminnow*	1	98.7	191						
Shimofuri Goby	3	559.7	68-72						
Splittail*							1	1.9	155
Striped Bass				5	334.2	79-312	82	139	56-320
Threadfin Shad	12	1352.9	46-88	66	4436	42-102	38	67.3	46-104
Wakasagi				2	123.8	75-82			
White Sturgeon*							1	1.5	454
Yellowfin Goby	10	1653.9	67-145	2	123.8	54-82			
<i>Crangon spp.*</i>				1	40.9		7	10.8	
<i>Exopalaemon spp.</i>	3	478.9		1	76.5		13	21.3	
<i>Maeotias spp.</i>				34	2152.3		4903	8313.8	

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Total	1829	326351.5		170	10672.3		5446	9247.7	
Total (fish only)	1826	325872.7	20-225	134	8402.6	42-312	523	901.9	42-454

TABLE 9. CPUE COMPARISON VALUES FOR GEAR TYPES IN VARIOUS HABITATS.

Shallow vs Channel Habitat					
Decker Island (n = 9)					
Gear Type	Mean CPUE	Std. Error		Wilcoxon Paired T-Test	
				Z value	P value
Beach Seine	36208.1	16123.1		2.5	0.01
Midwater Trawl	48.3	21.7			
Shallow vs Shallow Habitat					
Tule Red (n = 16)					
Gear Type	Mean CPUE	Std. Error		Wilcoxon Paired T-Test	
				Z value	P value
Lampara	525.2	183.5		3	0.003
Midwater Trawl	33.6	7.7			

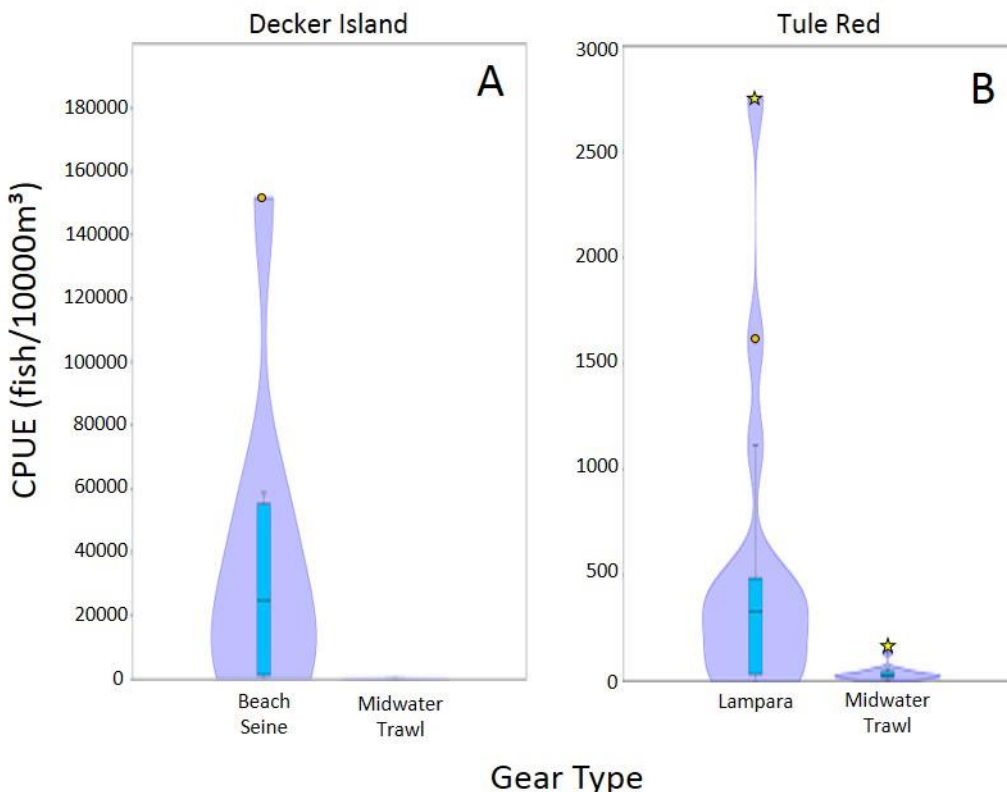


FIGURE 12 CPUE VIOLIN BOXPLOTS OF THE GEAR TYPES IN VARIOUS SAMPLING HABITATS. THE SHADED PURPLE AREAS OVERLAYING THE TRADITIONAL BOX PLOTS INDICATE PROBABILITY DENSITY. THE ORANGE CIRCLES REPRESENT VALUES OUTSIDE THE UPPER INNER FENCE AND YELLOW STARS REPRESENT VALUES 3 TIMES THE BOX HEIGHT FROM THE BOX.

The Kolmogorov-Smirnov test comparing fish sizes showed significant differences between all the gear types based on habitat sampled (Table 10), where fish size retention increased from the beach seine → lampara → midwater trawl (Figure 13). The beach seine caught a higher proportion of smaller fish primarily comprised of Mississippi Silversides and Rainwater Killifish (Figure 13, Figure 14). The lampara net caught a wide range of lengths for American Shad and Threadfin Shad, while the midwater trawl caught a wider range of lengths of American Shad, Longfin Smelt, Northern Anchovy, and Striped Bass (Figure 14).

TABLE 10. KOLMOGOROV-SMIRNOV COMPARISONS OF FISH FORK LENGTH SIZES BETWEEN GEAR TYPES IN THE FALL.

Beach Seine		Lampara	
N:	334	N:	187
D :	0.40781	p:	3.71E-18
Beach Seine		Midwater	
N:	334	N:	334
D :	0.58084	p:	2.43E-50
Lampara		Midwater	

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N:	187	N:	334
D :	0.25524	p:	2.27E-07

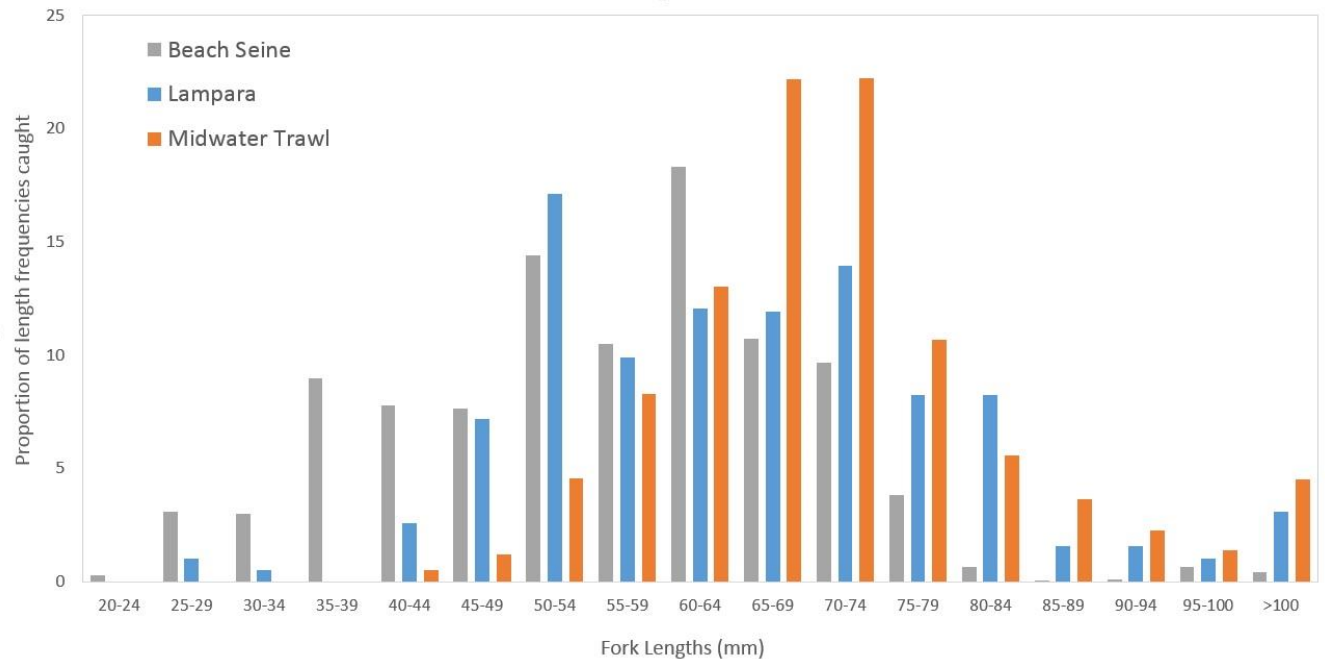


FIGURE 13. THE PROPORTION OF FORK LENGTH FREQUENCY CAUGHT BY EACH GEAR TYPE. FISH GREATER THAN 100 MM WERE NOT USED FOR LENGTH COMPARISONS BETWEEN GEAR TYPES, BUT ARE SHOWN HERE FOR ADDITIONAL INFORMATION.

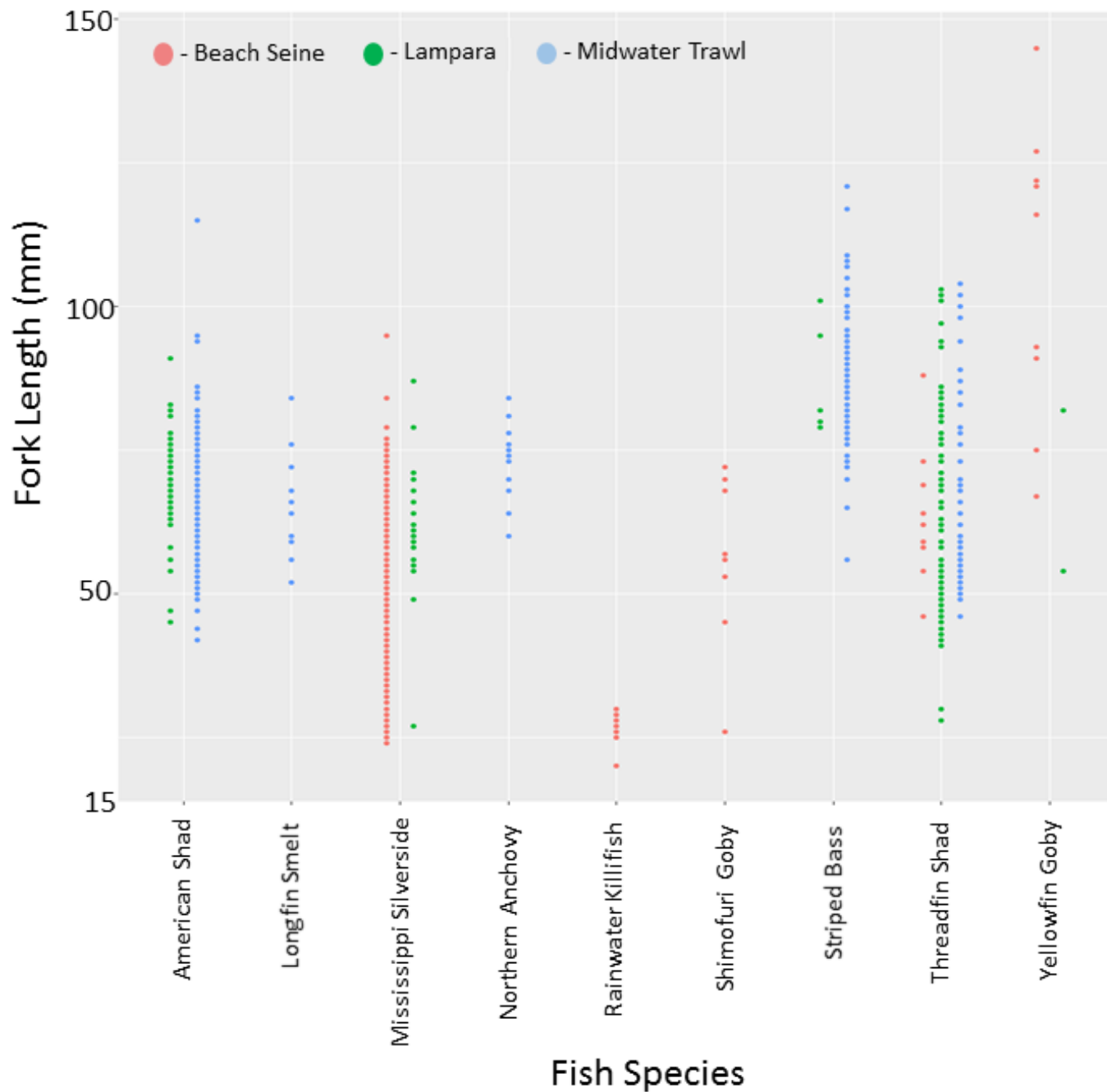


FIGURE 14. FORK LENGTHS FOR COMMON FISH CAPTURED BY THE BEACH SEINE, LAMPARA NET, AND MIDWATER TRAWL. EACH DOT REPRESENTS AN INDIVIDUAL FISH LENGTH CAUGHT BY EACH GEAR TYPE.

Fish composition between gear types differed outside Decker Island and Tule Red (Table 11). Each gear type caught a different array of fish species, but each gear type caught between 8-10 fish species (Table 8, Table 11). Month was also a predictor of differences in fish composition when the gears were compared in the shallow waters outside Tule Red. Both the beach seine and lampara net caught a higher abundance of Mississippi Silverside and Threadfin Shad (Figure 15).

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TABLE 11. RESULTS OF PERMANOVAS ON RELATIVE ABUNDANCE OF FISH SPECIES DURING THE FALL MIDWATER TRAWL SURVEY. ALL HABITAT COMPARISONS SHOW DIFFERENCES OF FISH COMPOSITION INFLUENCED BY GEAR TYPE.

PERMANOVA Channel Vs Shallow Water - Decker Island						
	Df	Sums of Sqs.	Mean Sqs	F-value	R ²	p-value
Gear.Type	1	2.8303	2.83032	18.1234	0.56837	0.01 **
Month	1	0.1644	0.16439	1.0527	0.03301	0.28
Temp	1	0.2466	0.2466	1.579	0.04952	0.19
SpC	1	0.0205	0.02054	0.1315	0.00412	0.89
Residuals	11	1.7179	0.15617		0.34497	
Total	15	4.9797			1	

PERMANOVA Shallow Vs Shallow Water – Tule Red						
	Df	Sums Of Sqs.	Mean Sqs	F-value	R ²	p-value
Gear.Type	1	0.8821	0.88206	4.7899	0.15024	0.01 **
Month	1	0.458	0.45798	2.487	0.07801	0.02 *
Temp	1	0.0083	0.00829	0.045	0.00141	0.97
SpC	1	0.2873	0.28726	1.5599	0.04893	0.15
Residuals	23	4.2354	0.18415		0.72141	
Total	27	5.871			1	

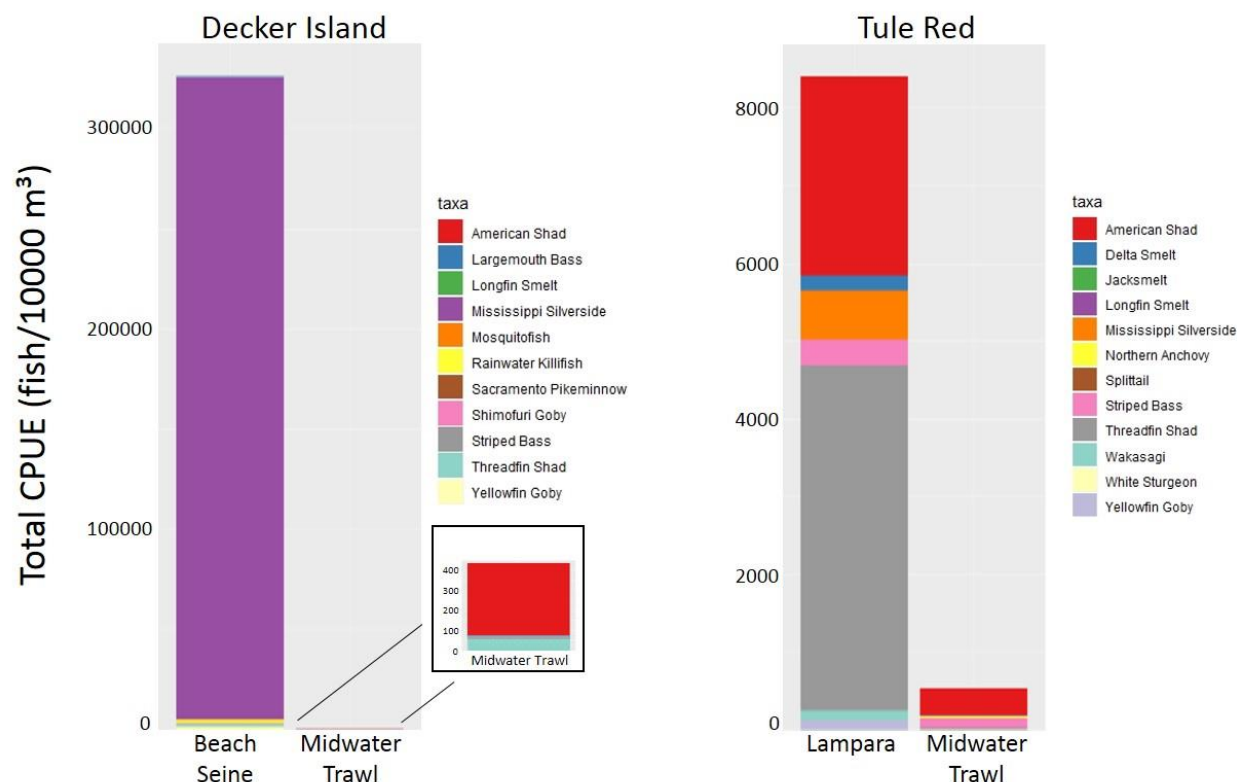


FIGURE 15. STACKED BAR CHART OF THE TOTAL CPUE OF FISH SPECIES CAUGHT BY EACH GEAR TYPE OUTSIDE FUTURE RESTORED TIDAL WETLANDS DURING FALL. AT DECKER ISLAND AN ENLARGED STACKED BAR GRAPH APPEARS TO THE RIGHT OF THE MIDWATER TRAWL STACKED BAR CHART TO SHOW WHICH FISH WERE CAUGHT.

Invertebrates

There were major differences in total mesozooplankton CPUE over time and space, with higher catches later in the spring, and higher catches further upstream (Figure 16, Table 12). Community composition also varied across both time and space. Both the PerMANOVA and NMDS demonstrate higher proportions of calanoid copepods later in the year, and higher proportions of Cladocera further upstream (Table 13,

Figure 17). Very low stress in the NMDS (0.07), and high R^2 in both the PerMANOVA (0.47) and GAMs (0.80 and 0.15) indicate these models provided a very good fit to the data.

The patterns in macrozooplankton CPUE and community composition were less reliable. There was a small, significant effect of distance from the Golden Gate in overall macrozooplankton CPUE, with higher catches further upstream (Figure 18, Table 14). There was no significant effect of time. Community composition also varied slightly with distance, but not with time. Both the PerMANOVA and NMDS showed a small, significant effect of distance, with more mollusks, annelids, and insects further upstream (Table 15, Figure 19), but no significant effect of time. Relatively high stress on the NMDS (0.21), and low R^2 on the PerMANOVA (0.09) indicate these models did not provide particularly good fits to the data. Overall, macrozooplankton were more variable than mesozooplankton, so additional years of data collection may be necessary before conclusions can be reached.

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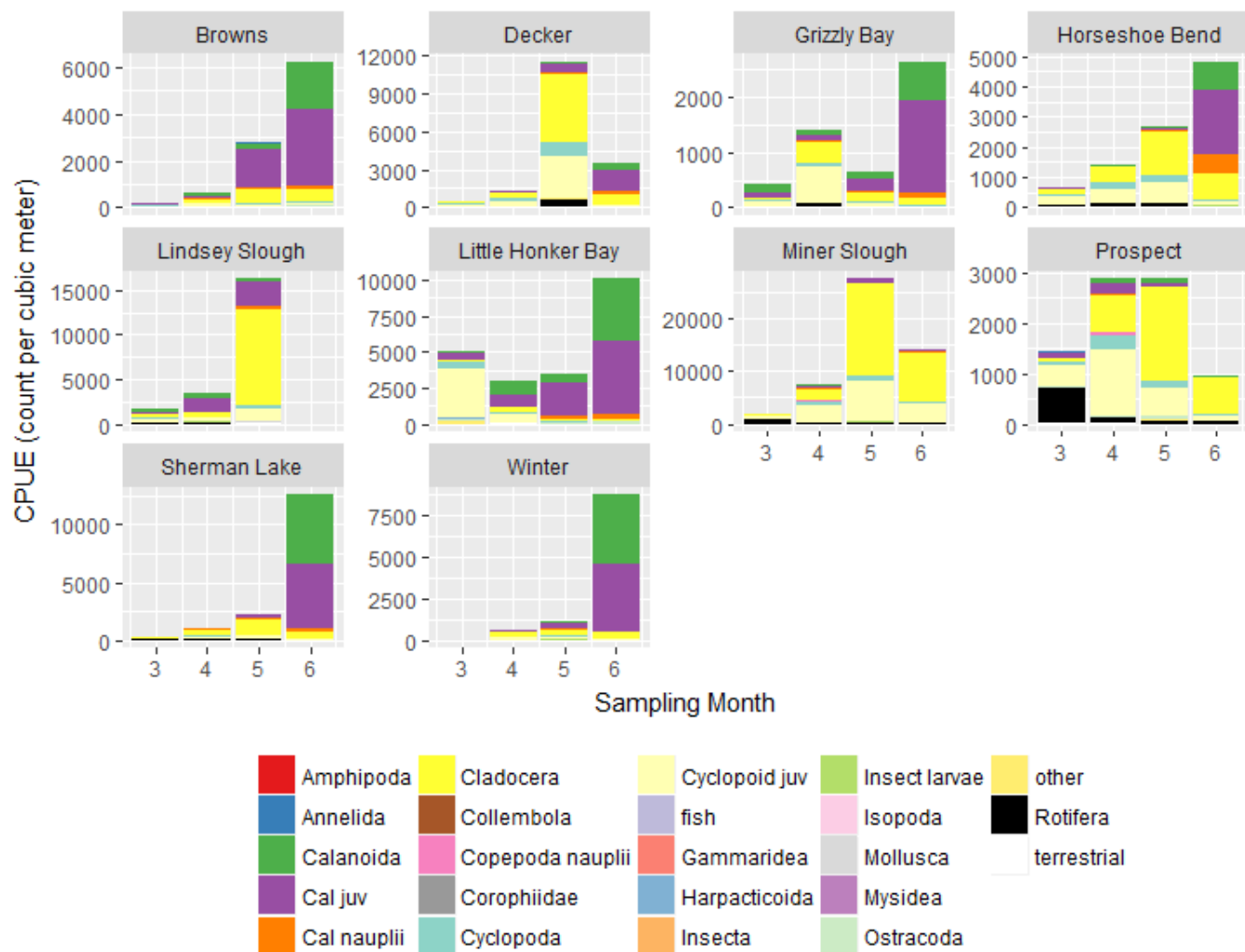


FIGURE 16. CPUE OF MAJOR ZOOPLANKTON TAXONOMIC GROUPS CAUGHT IN FRP TRAWLS IN EACH REGION OF THE ESTUARY, MARCH-JUNE, 2017.

TABLE 12. GENERALIZED LINEAR MODEL OF LOG TOTAL ZOOPLANKTON CATCH. THERE WAS A SIGNIFICANT EFFECT OF MONTH OF THE YEAR AND DISTANCE FROM THE GOLDEN GATE, WITH HIGHER ABUNDANCE LATER IN THE YEAR AND HIGHER ABUNDANCE IN FRESH WATER.

Term	Estimate	SE	t-value	p-value	
Intercept	2.278	0.918	2.438	0.017	*
Month	0.699	0.134	5.212	<0.001	*
Distance	0.024	0.007	3.441	0.001	*

TABLE 13. RESULTS OF A PERMANOVA ON RELATIVE ABUNDANCE OF MAJOR ZOOPLANKTON TAXONOMIC GROUPS. THERE WERE SIGNIFICANT DIFFERENCES OVER THE COURSE OF THE SPRING AND IN DIFFERENT REGIONS OF THE ESTUARY. OVERALL MODEL $R^2 = 0.473$, $F = 18.76$ ON 2 AND 38 DF, P -VALUE <0.0001 .

Term	DF	Sum of Squares	R^2	F-value	P-value
Month	1	1.28	0.23	13.04	0.001 *
Distance	1	0.62	0.11	6.33	0.004 *
Residuals	40	5.65	0.66		

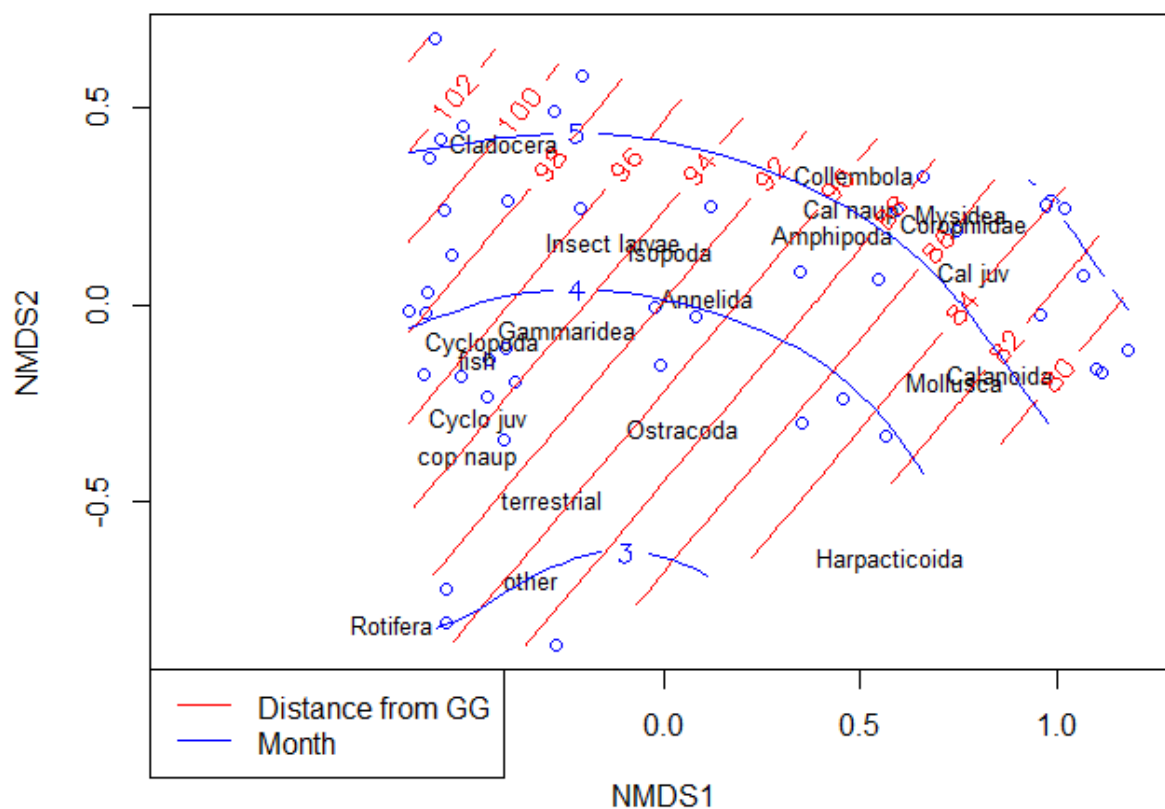


FIGURE 17. NMDS PLOT OF RELATIVE ABUNDANCE OF MAJOR ZOOPLANKTON TAXONOMIC GROUPS IN FRP SAMPLES TAKEN MONTHLY ACROSS THE DELTA AND SUISUN MARSH MARCH-JUNE 2017 (STRESS = 0.077). POINTS REPRESENT SAMPLES, TEXT REPRESENTS SPECIES. DISTANCE FROM THE GOLDEN GATE (IN KM) IS OVERLAID IN RED (GAM SIGNIFICANCE OF SMOOTHED TERMS $F = 0.797$, P -VALUE = 0.0162 ON 9 DEGREES OF FREEDOM, $R^2 = 0.152$), AND COLLECTION MONTH IS OVERLAID IN BLUE (GAM APPROXIMATE SIGNIFICANCE OF SMOOTHED TERMS F -VALUE = 11.06, P -VALUE <0.0001 , $R^2 = 0.713$).

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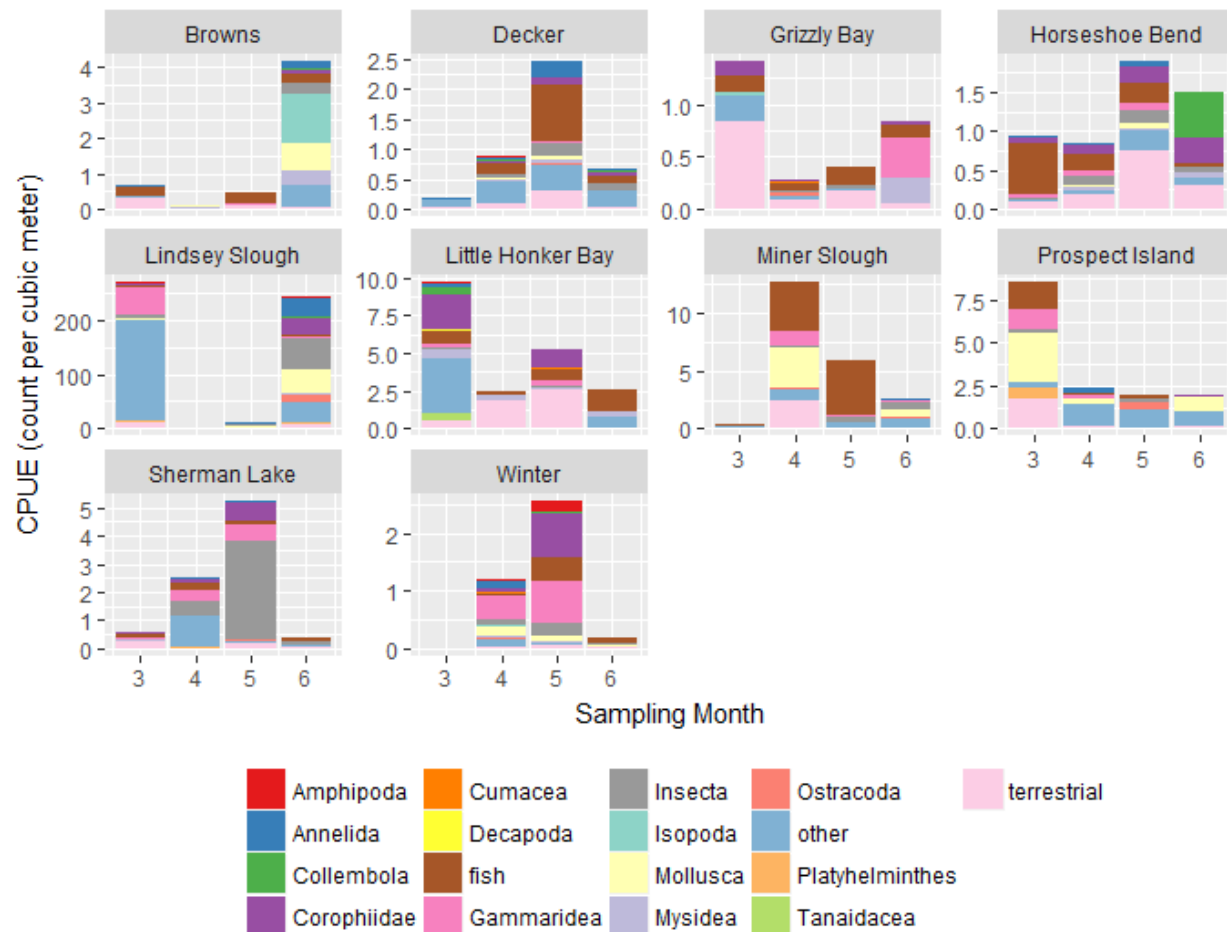


FIGURE 18. CPUE OF MAJOR MACROZOOPLANKTON TAXONOMIC GROUPS CAUGHT BY FRP IN MYSID NET TRAWLS, MARCH-JUNE, 2017.

TABLE 14. LINEAR MODEL OF LOG TOTAL MACROZOOPLANKTON CATCH. THERE WAS A SIGNIFICANT EFFECT OF DISTANCE FROM THE GOLDEN GATE, WITH HIGHER ABUNDANCE IN FRESH WATER, BUT NO SIGNIFICANT EFFECT OF MONTH OF THE YEAR. OVERALL MODEL $R^2 = 0.298$, $F = 9.91$ ON 2 AND 40 DF, $P\text{-VALUE} = 0.0003$.

Term	Estimate	SE	t-value	p-value
Intercept	-4.27	1.33	-3.21	0.003 *
Month	0.090	0.203	0.446	0.658
Distance	0.047	0.011	4.41	<0.0001 *

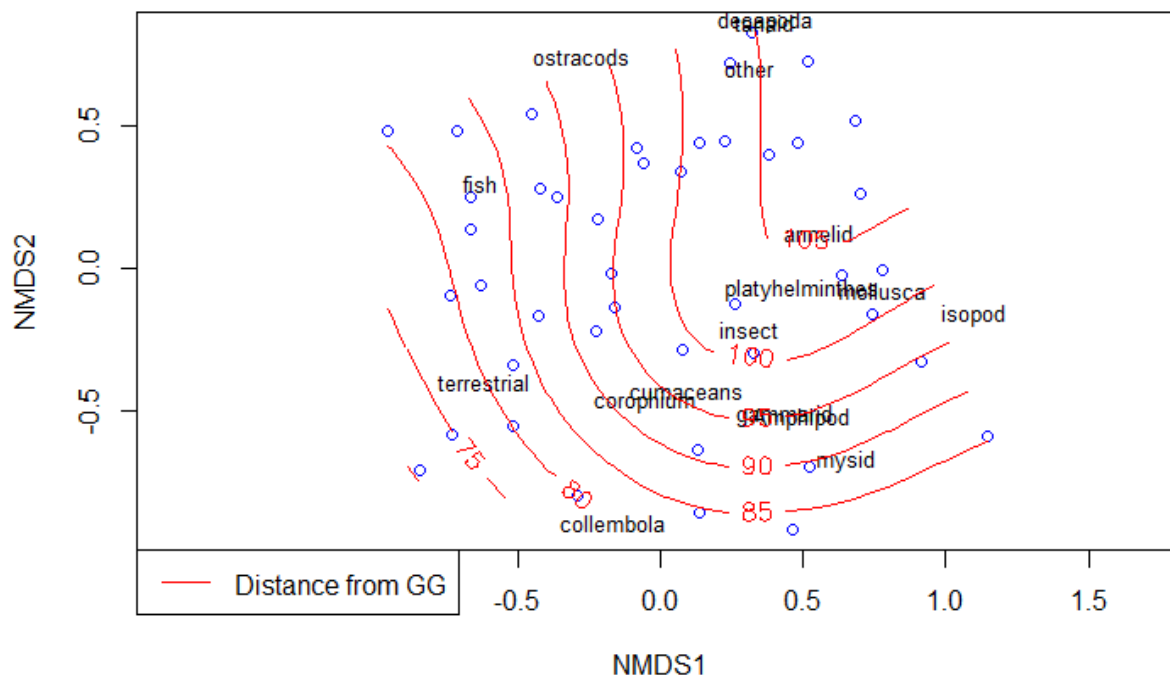


FIGURE 19. NMDS PLOT OF RELATIVE ABUNDANCE OF MAJOR MACROZOOPLANKTON TAXONOMIC GROUPS IN FRP MYSID SAMPLES TAKEN MONTHLY ACROSS THE DELTA AND SUISUN MARSH MARCH-JUNE 2017 (NMDS STRESS = 0.215). POINTS REPRESENT SAMPLES, TEXT REPRESENTS SPECIES. DISTANCE FROM THE GOLDEN GATE (IN KM) IS OVERLAID IN RED (GAM SIGNIFICANCE OF SMOOTHED TERMS $F = 2.089$, $P\text{-VALUE} = 0.0014$ ON 9 DEGREES OF FREEDOM, $R^2 = 0.31$), MONTH OF THE YEAR WAS NOT A SIGNIFICANT PREDICTOR VARIABLE IN EITHER THE PERMANOVA OR GAM, AND COULD NOT BE PLOTTED ON THE NMDS.

TABLE 15. RESULTS OF A PERMANOVA ON RELATIVE ABUNDANCE OF MAJOR MACROZOOPLANKTON TAXONOMIC GROUPS. THERE WAS A SIGNIFICANT EFFECT OF DISTANCE TO THE OUTLET OF THE ESTUARY, BUT NOT MONTH OF THE YEAR.

Term	DF	Sum of Squares	R ²	F-value	p-value
Month	1	0.313	0.033	1.51	0.18
Distance	1	0.862	0.09	4.17	0.002 *
Residuals	40	8.27	0.876		

Discussion

Fish

To date, shallow water sampling has never occurred simultaneously with the Summer Townet and Fall Midwater Trawl surveys. However, it comes as no surprise that fish abundance was higher in shallow water than in channel habitats. This may be because there are more fish and/or higher fish catch efficiency in shallow water habitat. Since many fish in the estuary are born between March and June (Meng and Matern 2001), catch was likely higher during June and July because the slow swimming

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speed of small fish makes them more susceptible to capture. Shallow water habitat is thought to provide better rearing habitat for certain fish species and this study supports that idea (Grimaldo et al. 2004; Sommer et al. 2005). In this study we observed the largest catches of juvenile Splittail and Sacramento Suckers occurring in June and July in shallow water habitat, where they were likely rearing (Kratville 2008; Moyle 2002; Sommer et al. 2004). Some of the other abundant fish caught in shallow water habitat that may be using it to rear were Threadfin Shad and Yellowfin Goby (Figure 11, Figure 15). In contrast, Striped Bass, *Tridentiger spp.*, and American Shad were more readily found within the channel and possibly rearing in this habitat. As other studies have demonstrated, some fish species favor different habitats and that idea is supported here (Grimaldo et al. 2004; Hagan and Able 2003).

During the summer, fish abundance, size, and composition were different when making comparisons between shallow water and channel gear types. Overall, the townet collected smaller fish than the beach seine and lampara net. The differences in fish composition between gear types is likely attributed to site depth, gear deployment, gear mesh size, and fish size. At sites where the depth exceeds 3.7 m, the lampara net does not sample the entire water column, as opposed to the townet. Larval/juvenile *Tridentiger spp.* and Striped Bass may have been located near the bottom of the channel and not effectively sampled by the lampara net (Contreras et al. 2017). The townet also has smaller cod end mesh that allowed many *Tridentiger spp.* and Striped Bass to be retained, as opposed to the lampara net. In contrast, the lampara caught more and larger pelagic fishes such as American Shad and Threadfin Shad, which may be due to how the gear is deployed. This idea was reiterated in Contreras et al. 2017 suggesting that the encircling deployment of the lampara may corral fish into the sampling area of the net. In addition, since the townet is an oblique trawl towed behind a boat, only a fraction of the volume sampled is surface water, and the boat may also scare a portion of pelagic fish away from entering the net (Bracciali et al. 2012; Claramunt et al. 2005; Contreras et al. 2017; Misund 1990)).

During the fall, fish abundance and composition differed between the lampara and midwater trawl in shallow habitat within Grizzly Bay, near Tule Red. Although some fish species, such as American Shad and Threadfin Shad, were regularly caught by both gears, fish composition differences appear to be driven by the relative abundance of these fish and more frequent catches of Striped Bass by the midwater trawl. Since the lampara net samples a smaller water volume than the midwater trawl, the CPUEs of fish were higher even though fewer fish were caught. Using the average volumes sampled by each gear, it is interesting to note that 1 fish caught in the lampara net \approx 30 fish caught in the midwater trawl. Therefore, it may appear that midwater trawl abundances are mistakenly low or that the higher abundances observed in the lampara net may be inflated. However, although the Striped Bass relative abundance was lower in the midwater trawl, this gear frequently captured them from September through December, while the lampara net only captured them in October. In addition, Longfin Smelt were exclusively caught by the midwater trawl from October to December, while the lampara net caught none during this time period even though both gears sampled in shallow water habitat. It may be that the faster towing speed of the midwater trawl makes Longfin Smelt more susceptible to this gear; however, this is purely speculation.

Overall, we found differences between sampling gear types. This suggests that although the different gear types typically sampled within three miles of one another, habitat and gear type influences the number and species of fish caught. Data collected from the channel does not characterize the shallow water habitat fish community. In order to determine what wetland benefits occur after restoration, shallow water sampling by the beach seine and lampara can provide useful fish data such as foraging or

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rearing patterns not observed by gear types trawling in the channel. A follow up study is slated for 2018 to continue these efforts and expand sampling to Ryer Island and Browns Island.

Within this study, the beach seine and lampara caught higher abundances of fishes overall. Although these two gear types performed well, the beach seine still caught more and smaller fish than the lampara net, probably due to differences in mesh size (Contreras et al. 2017). However, the lampara net is more robust and can sample more habitat types than the beach seine, which may be necessary to sample tidal wetlands. We expect many of the restored tidal wetlands to be dynamic and the lampara net's flexibility is an added bonus when choosing sampling gear types.

Invertebrates

We found clear patterns in mesozooplankton abundance and community composition near tidal wetlands across the estuary over the course of the spring. Zooplankton abundance was higher in freshwater, with a greater dominance of Cladocera (Figure 18, Figure 19). This is in keeping with IEP's zooplankton survey, which finds Cladocera most common upstream of the Confluence (Hennessy and Enderlein 2013), and higher zooplankton CPUE overall upstream of Suisun Bay (Hammock et al. 2017; Winder and Jassby 2011). This pattern has been hypothesized to be due to decreases in productivity caused by introduction of the brackish-water clam, *Potamocorbula amurensis* (Winder and Jassby 2011), and the predatory copepod *Acartiella sinensis* in freshwater (Contreras et al. 2017; Kayfetz and Kimmerer 2017). The increase in total CPUE and change in community composition from March through June is also supported by similar data collected by IEP. Zooplankton abundance in the estuary follows chlorophyll-*a* concentrations, with peak abundance in late spring or summer (Merz et al. 2016). Since its introduction, *Pseudodiaptomus forbesi* has become the most numerically dominant calanoid copepod in the low salinity zone (0.5-6 PSU) and freshwater reaches of the estuary, replacing *Eurytemora affinis* (Winder and Jassby 2011). The large increase in overall zooplankton CPUE found in our data was driven by the large increase in calanoid copepods, particularly *P. forebesii* (Figure 16), which usually experiences peak abundance in May or June, though this peak has shifted earlier since the introduction of *P. amurensis* (Merz et al. 2016).

Patterns in macrozooplankton abundance were less clear. Both overall CPUE and community composition were highly variable (Figure 18), so increased sample size may be necessary to make confident conclusions about overall patterns. We found no trends in CPUE or community composition over time, contrary to trends in IEP's Zooplankton Survey data, which generally finds mysid abundance to peak in late spring or early summer (Hennessy and Enderlein 2013). A study of mysids in Suisun marsh found the peak abundance varies by species, with *Neomysis kadiakensis* peaking early, and *Acanthomysis bowmani*, followed by *N. mercedis* later in the season (Carlson and Matern 2000).

It is worth noting, however, that there are few data sets on macroinvertebrates with which we can compare our data. The Zooplankton Survey only enumerates mysids and amphipods, not insects, isopods, or other crustaceans. Therefore, it is difficult to determine the extent to which our results match that found by other researchers. One of the few studies looking at the macrozooplankton community as a whole also found very high spatial and temporal variability, with seasonal peaks in summer or fall, depending on region of the estuary and salinity (Gewant and Bollens 2005).

Part 2. Timing of Food Web Sampling

Introduction

FRP has spent the past several years refining the gear types necessary to sample macroinvertebrates and zooplankton in tidal wetlands. However, the extreme spatial and temporal variability in aquatic invertebrate sampling often makes it difficult to extract patterns from long-term datasets. Because invertebrate samples are extremely labor-intensive to process, FRP needs to identify the most efficient sampling replication, sampling frequency, spatial distribution, and temporal distribution.

Mesozooplankton are recognized as the largest component of Delta Smelt diets (Slater and Baxter 2014) and a significant component of salmon diets (Sommer et al. 2001) but many zooplankton exhibit tidal and diurnal vertical migrations (Kimmerer et al. 1998, 2002, Burks et al. 2002). By sampling during times when fish of concern are most active, we can characterize zooplankton most available for fish consumption. However, salmon are most active at night (Wilder and Ingram 2006, Plumb et al. 2016), while Delta Smelt are most active during the day (Young et al. 2004, Hasenbein et al. 2013). Though Delta Smelt feed chiefly during daylight hours, they have been documented feeding on more adult *Pseudodiaptomus forbesi* than would be expected given the relative abundance of this species in daytime zooplankton samples (Slater and Baxter 2014). Feeding during dawn and dusk, when adult *P. forbesi* and other copepods have migrated to the surface, may explain part of this discrepancy. Furthermore, when trying to characterize export of production from the wetland, we may be missing important components of the community that enter the pelagic food web at different times of day (Dean et al. 2005; Kimmerer et al. 2014).

Because epifaunal invertebrates are a smaller percentage of smelt diets (Slater and Baxter 2014) and are less mobile than zooplankton, it may only be necessary to sample these macroinvertebrates once or twice per year. If sampling is limited, we want to determine what time of year has greatest overlap between listed fish species and their food supply.

Research Questions

1. When during the daily or tidal cycle should zooplankton sampling occur?
 - a. Can we use daytime samples to infer total zooplankton abundance?
2. When during the year should macroinvertebrate sampling occur?

Methods

Tidal and diel timing

To determine whether we need to sample zooplankton during both day and night, we conducted an intensive comparison of zooplankton CPUE and community composition around Decker Island and Horseshoe Bend. Once during June, at the peak of vertical migrator *P. forbesi* abundance (Hennessy and Enderlein 2013; Merz et al. 2016), we collected a series of three replicate samples throughout the day and night at different depths (Table 16).

Sampling occurred approximately every four hours from 9:00am on June 26th, to 6:00am on June 27th. Sampling bouts occurred slightly closer together during the night to ensure at least four sampling bouts

could occur in relative darkness. Because of the relative time of sunset and sunrise, the “High Slack Day” sampling period overlapped with sunset, and the “High Slack Night” sampling period was slightly before high slack to avoid overlapping with sunrise (Figure 21).

Three sampling stations were sampled during each sampling bout, and each sampling station included one wetland trawl (along the edge of vegetation in 3-10 ft of water) and one or two channel trawls (center of channel in 20-30 ft of water). Channel stations included concurrent surface and benthic trawls during daylight, and surface trawls only during the night. We also took oblique trawls during the first sampling bout (mid ebb day), but these trawls were discontinued due to time constraints. Wetland stations were surface trawls only throughout. All trawls were 5-minutes long, against the tide when possible. Some of the trawls were with the tide due to high winds, and two trawls were cut to four minutes due to hazardous conditions.

Samples were rinsed in their entirety into jars, preserved in 70% EtOH, and identified in the lab to the lowest ecologically relevant taxa (see Lab Methods, Invertebrates, below).

Water quality (pH, DO, temperature, conductivity, chlorophyll florescence, and turbidity) was collected using a YSI 6600 at the bottom, middle, and top of the water column at each station. A smaller handheld sonde (YSI Proplus, pH, DO, temperature, and conductivity in surface water only) was used to verify YSI 6600 readings during the first five sampling bouts. Turbidity in the surface water was verified using a portable turbidity meter.



FIGURE 20. LOCATION OF SAMPLING STATIONS IN HORSESHOE BEND. EACH STATION (REP1, REP2, AND REP3) WAS SAMPLED ONCE PER SAMPLING PERIOD. STATIONS WERE INCLUDED AS BLOCKING VARIABLES IN MODELS OF CATCH AND COMMUNITY COMPOSITION.

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TABLE 16. SAMPLE SIZE FOR EACH SAMPLE TYPE AT EACH TIME PERIOD.

	Day				Night			
	Mid ebb	Low slack	Mid flood	High slack	Mid ebb	Low slack	Mid flood	High Slack
Sample type	9:45	12:45	16:15	19:30	22:15	1:00	2:45	14:22
Benthic channel	3	3	3	3				
Surface channel	3	3	3	3	3	3	3	3
Wetland	3	3	3	3	3	3	3	3
Total trawls: 72								

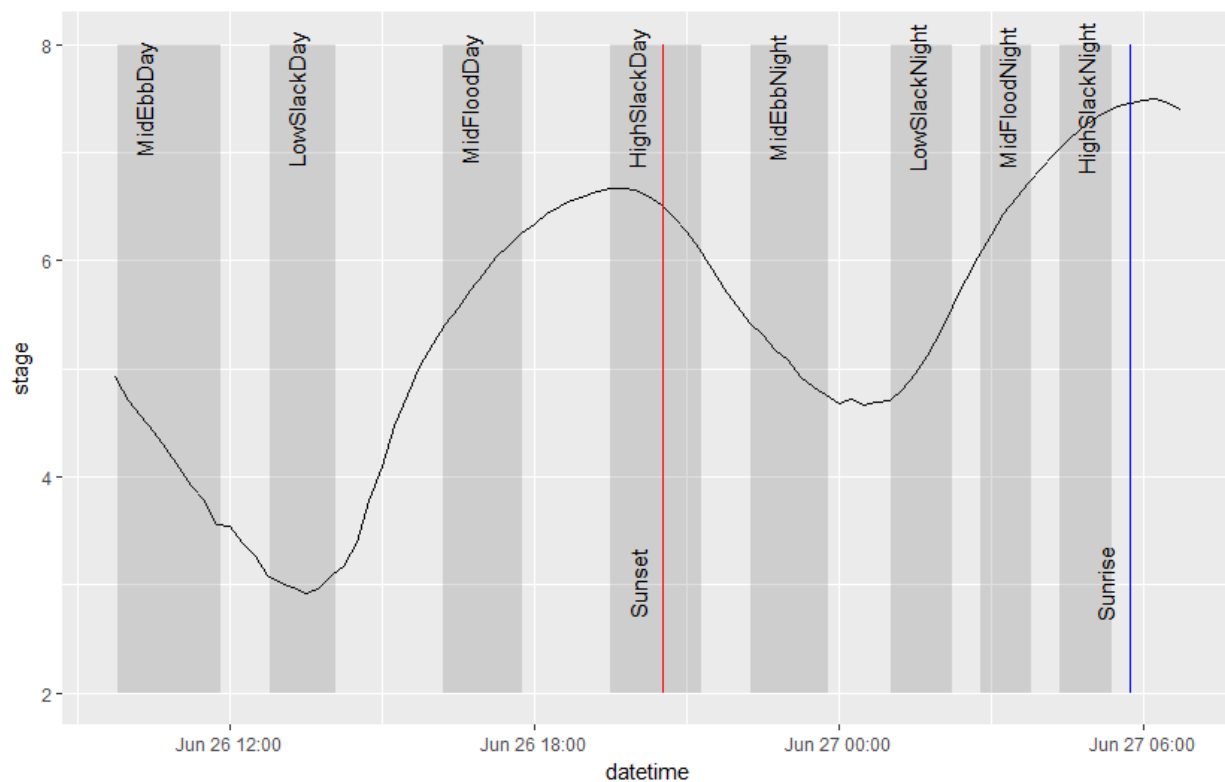


FIGURE 21. TIDAL STAGE VERSUS TIME FOR THE RIO VISTA USGS TIDE GAUGE (SITE NUMBER 11455420, DATA AVAILABLE: [HTTPS://WATERDATA.USGS.GOV/NWIS/INVENTORY?AGENCY_CODE=USGS&SITE_NO=11455420](https://waterdata.usgs.gov/nwis/inventory?agency_code=USGS&site_no=11455420)). GRAY PERIODS INDICATE WHEN SAMPLING BOUTS OCCURRED.

Analysis Methods

To analyze the effect of tidal stage and time of day, we subset the data to just include surface trawls, since benthic trawls were not collected at night. To test whether benthic trawls had higher abundance than surface trawls, we subset the data to just include daytime trawls.

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For each subset of the data, we performed a series of linear models on the log-transformed total zooplankton CPUE using the predictor variables listed in (Table 17 or 18, depending on the subset). We ranked all possible models using Akaike’s Information Criterion, corrected for small sample sizes (AICc), to choose which predictor variables to use in the final model using the R package MuMIN (Barton 2018). We also performed a Permutational Multivariate Analysis of Variance (PERMANOVA) using the “adonis” function from the R package vegan (Oksanen et al. 2016) to see whether the same predictor variables would have an effect on community composition.

Because calanoid copepods, specifically *P. forbesi*, was the numerically dominant taxonomic group in most of the samples, we repeated the analysis of log-transformed CPUE on the dataset of just the adult calanoid copepods, and on the dataset of everything besides calanoid copepods. This allowed us to see whether the evidence for vertical migration in *P. forbesi* extended to other zooplankton taxa.

TABLE 17. PREDICTOR VARIABLES FOR DAY/NIGHT COMPARISON MODELS. (USING DATA FROM SURFACE TOWS ONLY).

Variable	Variable type	Description	Interpretation
Tide	Categorical	Tidal stage at which sampling occurred: High Slack, Low Slack, Mid Ebb, or Mid Flood	Tidally-driven vertical migration
Day/Night	Categorical	Whether sample was collected during the day or at night.	Diurnal vertical migration
Wetland/Channel	Categorical	Whether the sample was collected in shallow water (< 3 m) adjacent to the wetland, or in the center of the channel (> 6 m deep).	Different abundances in shallow water versus the deep channel.
Day/Night* Tide interaction	Interaction		The effect of Day/Night is different at different tidal stages
Day/Night* Channel interaction	Interaction		The effect of Day/Night is different in the channel than the wetland.
Station	Categorical	Position along horseshoe bend (see Figure 20)	Blocking variable

TABLE 18. PREDICTOR VARIABLES FOR SURFACE/BENTHIC COMPARISON MODELS. (USING DATA FROM DAYTIME TOWS ONLY).

Variable	Variable type	Description	Interpretation
Tide	Categorical	Tidal stage at which sampling occurred: High Slack, Low Slack, Mid Ebb, or Mid Flood	Tidally-driven vertical migration
Surface/benthic	Categorical	Whether sample was collected from the surface of the channel or the bottom of the channel	Differing abundance at the top versus bottom of the water column
Wetland/Channel	Categorical	Whether the sample was collected in shallow water (< 3 m) adjacent to the wetland, or in the center of the channel (> 6 m deep).	Different abundances in shallow water versus the deep channel.
Tide* Surface/benthic interaction	Interaction		The effect of the tide is different at the top of the water column than the bottom
Station	Categorical	Position along horseshoe bend (see Figure 20)	Blocking variable

Intra-annual timing

To refine target sampling dates for future years, Decker Island was sampled for a broad suite of macroinvertebrates five times over the course of winter and spring (January 25, February 14, March 30, May 8, and June 5). Methods followed those outlined in *Part 3. Sample size and variability of food web data*, below. At each sampling time point, we collected sweep net samples, zooplankton tows, mysid tows, and neuston tows at or near the culvert providing muted tidal inflow to Decker Island, with three samples per habitat type.

We tested the fit of linear and quadratic equations of long-transformed CPUE of macroinvertebrates (excluding mesozooplankton) for each gear type to see when biomass peaks. A linear model suggests that invertebrate abundance increases steadily over the course of the spring, peaking sometime after sampling ended for the year. A quadratic model would suggest that abundance peaked sometime during our spring sampling period. Relative model fit was tested by comparing AICc and coefficient of determination (R^2) for the two models.

To show when at-risk fish abundance has the greatest overlap with macroinvertebrate abundance, we used data from the CDFW Spring Kodiak Trawl (SKT) survey for abundance of adult Delta Smelt at the

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station nearest Decker Island (station 706; data available: <https://www.wildlife.ca.gov/Conservation/Delta/Spring-Kodiak-Trawl>) and the USFWS Chipps Island Trawl for abundance of Chinook Salmon smolts moving through the Delta (data available: https://www.fws.gov/lodi/juvenile_fish_monitoring_program/jfmp_index.htm). Adult Delta Smelt and Chinook Salmon smolts were chosen because these are the life stages most likely to prey on macroinvertebrates (mysids, amphipods, and insects). We averaged catch of fishes in these two surveys for the past five years (2012-2017), and calculated the percentage of yearly catch that fell in each month. These data were plotted to see which month had the greatest total catch of fish of interest, with the best-fitting model of macroinvertebrate abundance overlaid.

Results

Tidal and diel differences

We found strong differences in total catch in different times of day and different stages of tide. The top model of log-transformed total zooplankton catch (surface samples only), included Day/Night, Station, Tide, and the interaction between Day/Night and Tide (Table 19). There was significantly higher catch at high slack tide during the day than the other tidal stages during the day, but all tide stages had equally high catch at night (Figure 22). AICc model selection did not support inclusion of Wetland/Channel as a predictor variable.

PerMANOVA also indicated significant differences in community composition at different times of day and different stages of the tide. NMDS plots show separation of the Day/Night and Tide sample groups, driven by the relative abundance of calanoid copepods (Table 20, Figure 23A, B). However, there was no significant difference in community composition in channel versus wetland habitat (Table 20, Figure 23C).

Because of the dominance of calanoid copepods, and previous research on vertical migration in *P. forbesi* (the dominant calanoid in our samples), we re-ran the analysis of log-transformed CPUE on just the adult calanoid copepods, and a separate model of the data set with no calanoid copepods. For adult calanoids, AICc model selection supported the same top model as the overall zooplankton model, with the same general trends in abundance: higher catch at night and higher catch at slack tide (Figure 24, Table 21). When calanoid copepods were removed from the analysis, the top model chosen via AICc only supported Day/Night and Station (Table 22), with significantly higher catches at night (Figure 26). Neither Tide nor Wetland/Channel were supported. The lack of support for tide in the model without calanoids indicates that many zooplankton exhibited diel vertical migration, but calanoid copepods were driving the tidal vertical migration patterns.

We also found a slight trend towards higher catch in benthic samples when compared with surface samples during the day, though abundance was highest at High Slack tide at all water depths (Figure 266). The top model of log-transformed total zooplankton catch (daytime samples only), included Surface/Benthic, Station and Tide (Table 23). There was no significant effect of Channel/Wetland and there was no interaction between Tide and Surface/Benthic.

While the effect on overall abundance was relatively small, PerMANOVA results indicated significant differences in community composition between surface and benthic samples, and between different

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stages of the tide. NMDS plots show separation of the Surface/Benthic and Tide sample groups, driven by the relative abundance of calanoid copepods (Table 24, Figure 27A, B). Unlike data from the surface samples, there was also a trend towards a significant effect of Channel/Wetland (Table 24, Figure 27C).

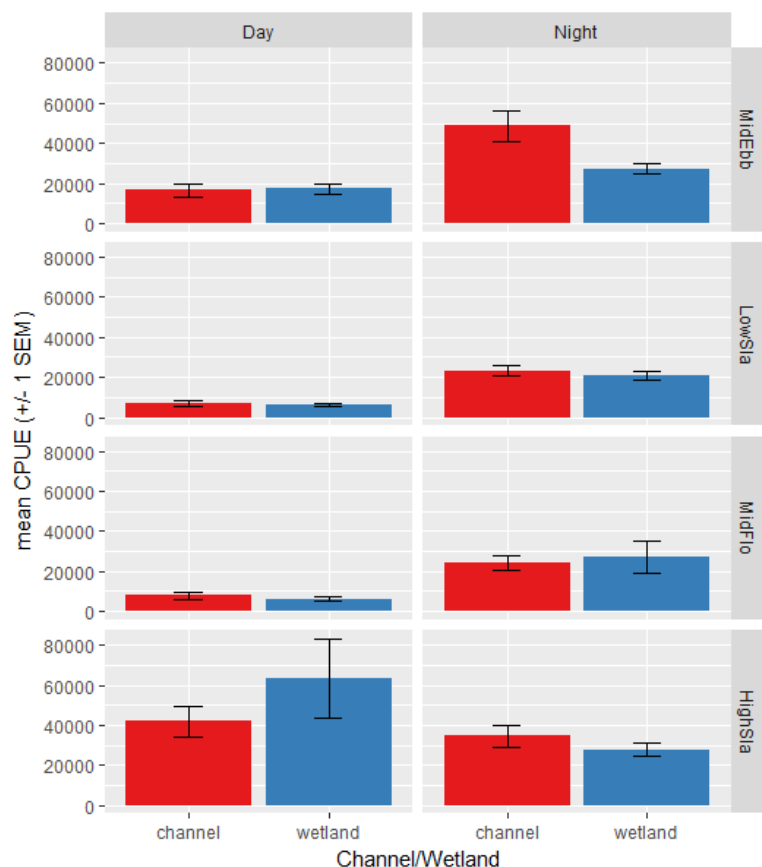


FIGURE 22. MEAN ZOOPLANKTON CPUE OF SURFACE SAMPLES AT EACH TIDE AND TIME OF DAY (MEAN OF THE THREE SAMPLING STATIONS). ALL TAXA HAVE BEEN COMBINED IN THIS GRAPH, BUT NOTE THAT OVER 90% OF THE ABUNDANCE WAS JUVENILE OR ADULT CALANOID COPEPODS. TIDAL STAGE ABBREVIATIONS: MIDEBB = MID EBB TIDE, LOWSLA = LOW SLACK TIDE, MIDFLO = MID FLOOD TIDE, HIGHSLA = HIGH SLACK TIDE.

TABLE 19. COEFFICIENTS OF THE TOP MODEL OF LOG TOTAL CPUE FOR DAYTIME SAMPLES. THE TERMS SUPPORTED BY THE MODEL SELECTION PROCESS WERE DAY/NIGHT, STATION, AND TIDE, PLUS THE INTERACTION OF DAY/NIGHT AND TIDE. WETLAND/CHANNEL WAS NOT SUPPORTED. ADJUSTED R-SQUARED = 0.739, OVERALL F-STATISTIC 15.81 ON 9 AND 38 DF, P-VALUE <0.0001.

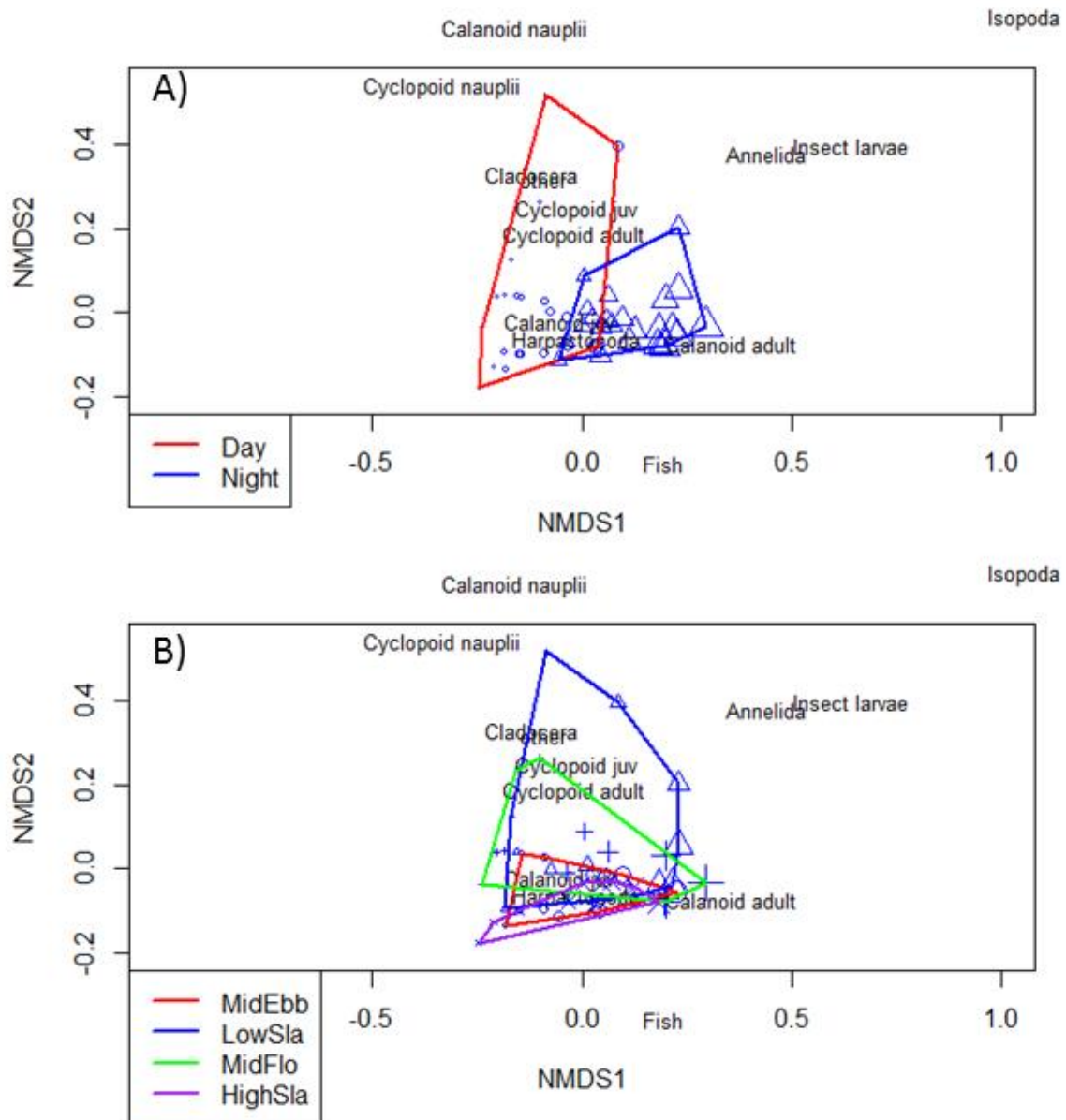
Term	Estimate	SE	t-value	p-value
Intercept - Day,				
HighSlack, Rep1	10.860	0.203	53.473	<0.0001 *
Night	-0.354	0.257	-1.379	0.176

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Low Slack	-1.919	0.257	-7.471	<0.0001	*
Mid Ebb	-1.035	0.257	-4.027	0.0002	*
Mid Flood	-1.937	0.257	-7.54	<0.0001	*
Rep2	-0.004	0.157	-0.026	0.979	
Rep3	-0.692	0.157	-4.396	<0.0001	*
Night*LowSlack	1.603	0.363	4.413	<0.0001	*
Night*MidEbb	1.210	0.363	3.33	0.0019	*
Night*MidFlood	1.641	0.363	4.517	<0.0001	*

TABLE 20. RESULTS OF A PERMANOVA ON RELATIVE ABUNDANCE OF MAJOR TAXA IN ALL THE SURFACE SAMPLES. RESULTS SHOWED A SIGNIFICANT EFFECT OF TIDE AND DAY/NIGHT, BUT NOT CHANNEL/WETLAND.

Term	DF	Sums of Sqs.	Mean Sqs.	f-value	R ²	p-value	
Day/Night	1	0.163	0.164	26.417	0.332	0.001	*
Tide	3	0.068	0.022	3.667	0.138	0.005	*
Channel/Wetland	1	0.001	0.001	0.207	0.003	0.845	
Residuals	42	0.260	0.006	0.527			
Total	47	0.493	1				



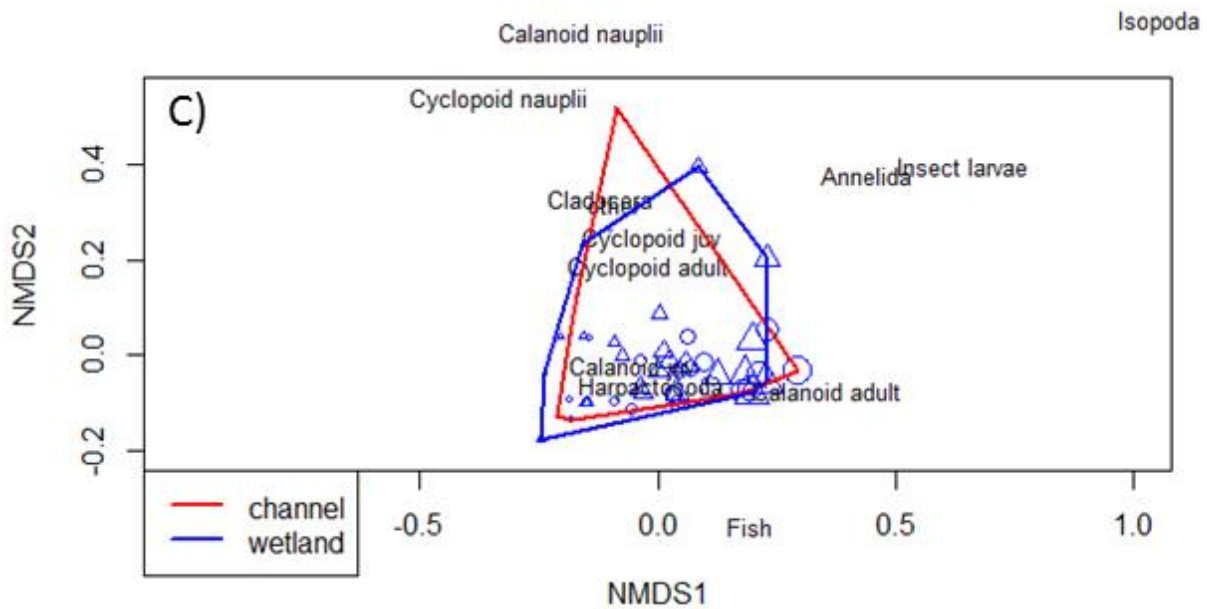


FIGURE 23. NMDS PLOT OF RELATIVE ABUNDANCE OF ZOOPLANKTON IN ALL SURFACE SAMPLES FROM THE 24 HOUR STUDY (STRESS = 0.0379). POINTS REPRESENT SAMPLES, TEXT REPRESENT SPECIES. SAMPLE POINT SIZE VARIES BY THE PROPORTION OF ADULT CALANOID COPEPODS A) NMDS PLOT WITH HULLS AROUND DAY AND NIGHT SAMPLES. PERMANOVA SUPPORTS THESE GROUPS BEING SIGNIFICANTLY DIFFERENT. B) NMDS PLOT WITH HULLS AROUND SAMPLES FROM DIFFERENT TIDAL STAGES. PERMANOVA SUPPORTS THESE GROUPS BEING SIGNIFICANTLY DIFFERENT. C) NMDS PLOT WITH HULLS AROUND CHANNEL AND WETLAND SAMPLE. THESE GROUPS ARE NOT SIGNIFICANTLY DIFFERENT. SEE TABLE 20 FOR PERMANOVA RESULTS.

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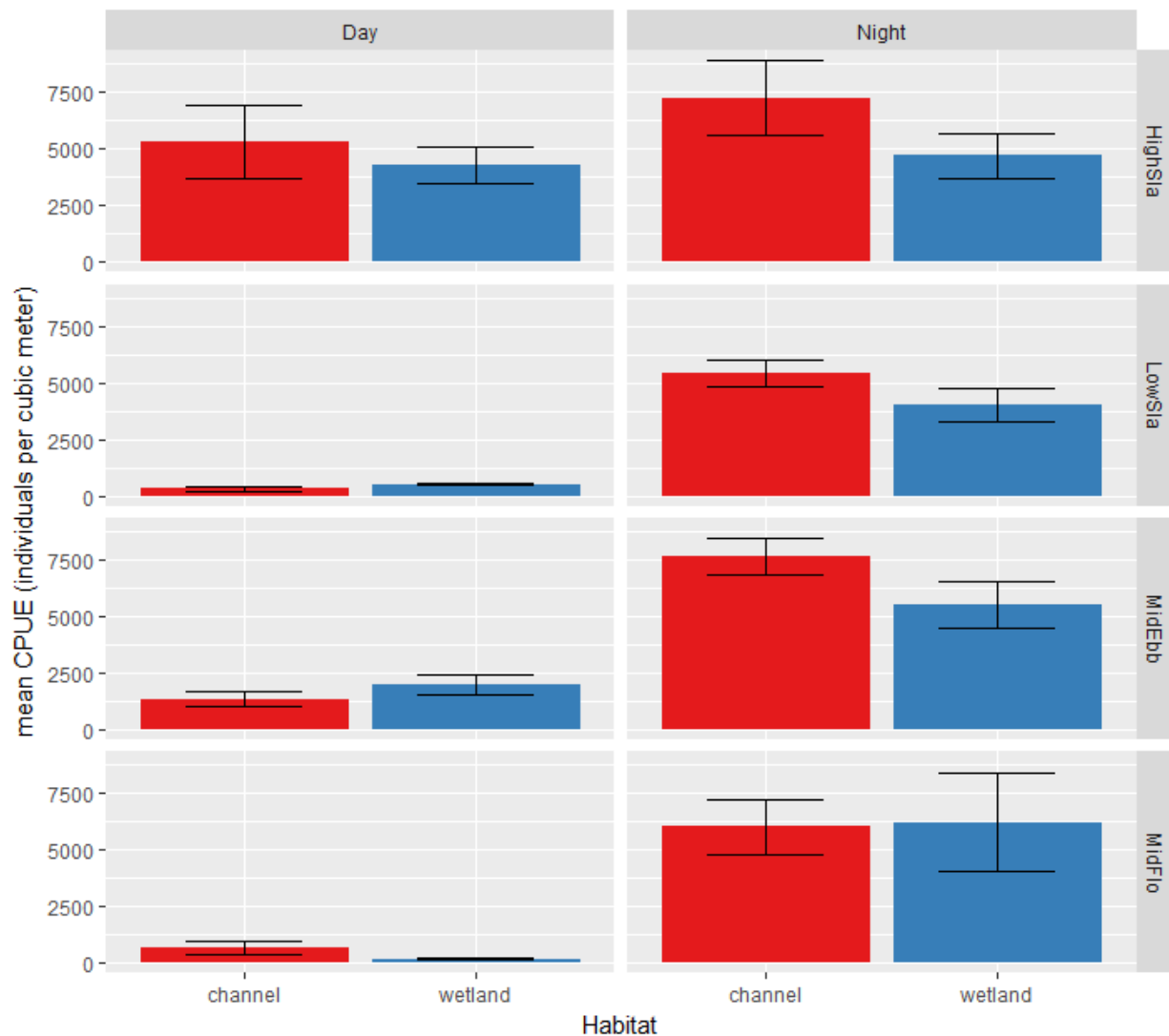


FIGURE 24. MEAN (+/- 1 SEM) OF THE ADULT CALANOID COPEPOD CPUE BY HABITAT, TIME OF DAY, AND TIDAL STAGE. SEE TABLE 21 FOR SIGNIFICANT DIFFERENCES.

TABLE 21. COEFFICIENTS OF THE TOP MODEL OF LOG CALANOID COPEPOD CPUE FOR SURFACE SAMPLES. THE TERMS SUPPORTED BY THE MODEL SELECTION PROCESS WERE THE SAME AS THE TOTAL CPUE MODEL: DAY/NIGHT, STATION, AND TIDE, PLUS THE INTERACTION OF DAY/NIGHT AND TIDE). WETLAND/CHANNEL WAS NOT SUPPORTED. ADJUSTED R-SQUARED = 0.799, OVERALL F-STATISTIC 21.74 ON 9 AND 38 DF, P-VALUE <0.0001.

Term	Estimate	SE	t-value	p-value	
Intercept - Day,					
High Slack, Rep1	8.639	0.292	29.603	<0.0001	*
Night	0.278	0.369	0.752	0.457	
Low Slack	-2.469	0.369	-6.687	<0.0001	*

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Mid Ebb	-1.010	0.369	-2.979	0.005	*
Mid Flood	-2.780	0.369	-7.531	<0.0001	*
Rep2	-0.149	0.226	-0.659	0.514	
Rep3	-1.000	0.226	-4.425	<0.0001	*
Night*LowSlack	2.314	0.522	4.432	<0.0001	*
Night*MidEbb	1.275	0.522	2.441	0.0194	*
Night*MidFlood	2.653	0.522	5.081	<0.0001	*

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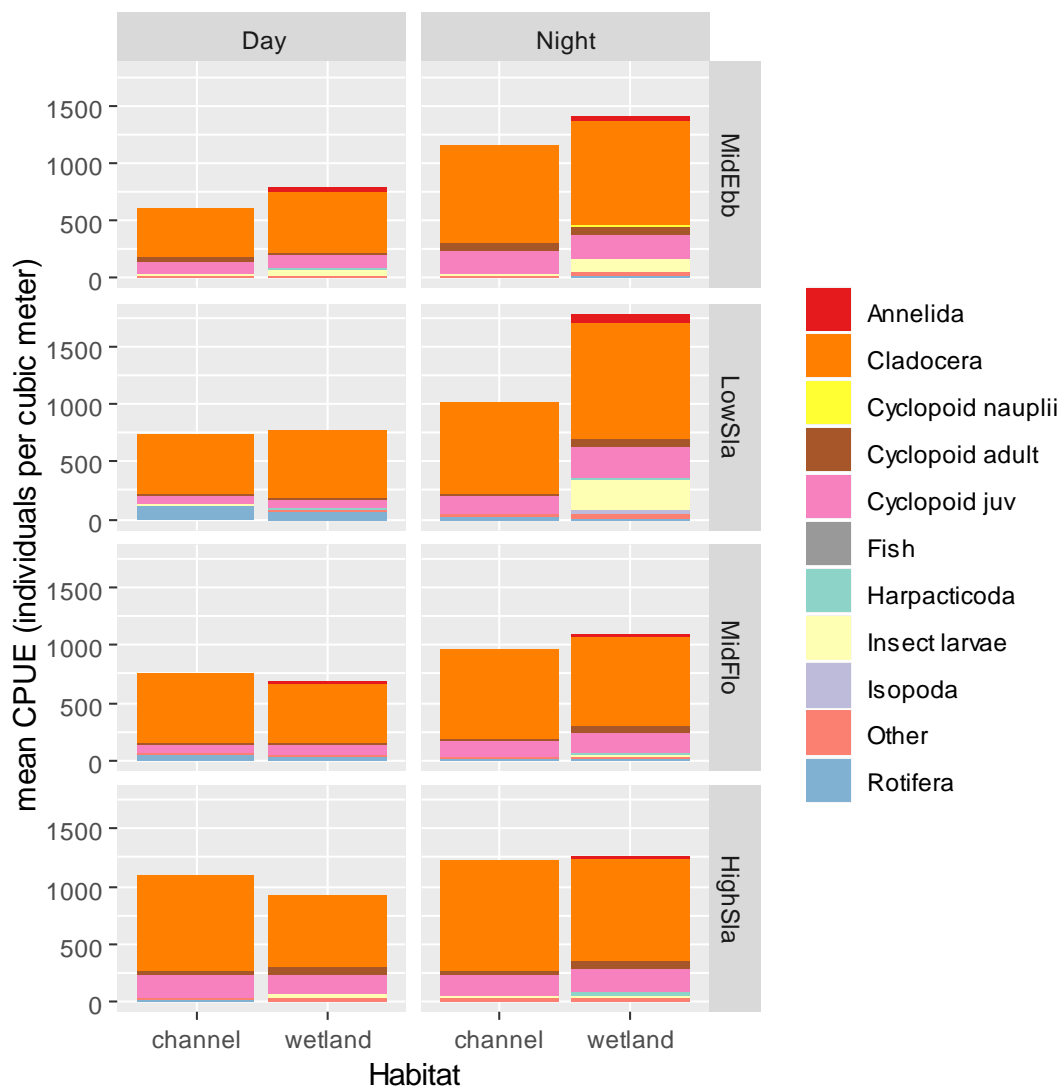


FIGURE 25. MEAN ZOOPLANKTON CPUE OF SURFACE SAMPLES WITH CALANOID COPEPODS REMOVED.

TABLE 22. COEFFICIENTS OF THE TOP MODEL OF LOG ZOOPLANKTON CPUE FOR SURFACE SAMPLES WITH CALANOID COPEPODS REMOVED. THE ONLY TERMS SUPPORTED BY THE MODEL SELECTION PROCESS WERE DAY/NIGHT AND STATION. WETLAND/CHANNEL AND TIDE WERE NOT SUPPORTED. ADJUSTED R-SQUARED = 0.387, OVERALL F-STATISTIC 10.91 ON 3 AND 44 DF, P-VALUE <0.0001.

Term	Estimate	SE	t-value	p-value	
Intercept - Rep1,					
Day	6.454	0.088	72.997	<0.0001	*
Night	0.466	0.088	5.268	<0.0001	*
Rep2	0.222	0.108	2.052	0.046	*
Rep3	0.194	0.108	1.791	0.080	.

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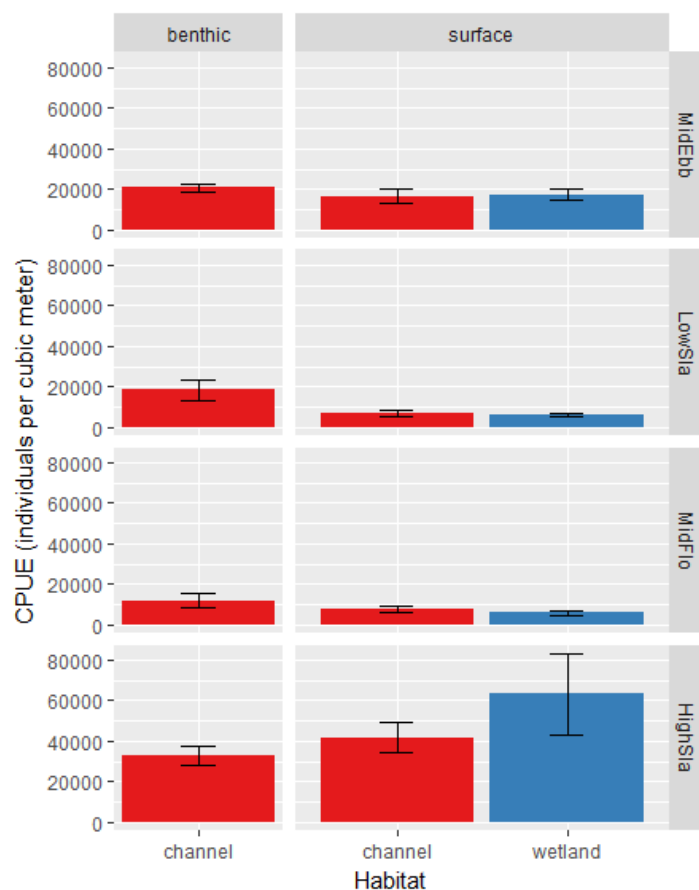


FIGURE 26. MEAN ZOOPLANKTON CPUE (+/- 1 SEM) OF DAYTIME SAMPLES FOR EACH HABITAT TYPE AT EACH TIDE. SEE TABLE 23 FOR SIGNIFICANT DIFFERENCES.

TABLE 23. COEFFICIENTS OF THE TOP MODEL OF LOG TOTAL CPUE FOR DAYTIME SAMPLES. THE TERMS SUPPORTED BY THE MODEL SELECTION PROCESS WERE STATION, SURFACE VERSUS BENTHIC, AND TIDE. WETLAND VERSUS CHANNEL AND ANY INTERACTION TERMS WERE NOT SUPPORTED. ADJUSTED R-SQUARED = 0.705, OVERALL F-STATISTIC 14.92 ON 6 AND 29 DF, P-VALUE <0.0001.

Term	Estimate	SE	t-value	p-value	
Intercept - Rep1, Benthic,					
HighSlack	10.726	0.237	45.235	<0.001	*
Rep2	0.524	0.205	2.550	0.016	*
Rep3	-0.461	0.205	-2.244	0.032	*
Surface	-0.334	0.178	-1.875	0.070	.

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Low Slack	-1.529	0.237	-6.448	<0.001	*
Mid Ebb	-0.824	0.237	-3.476	0.002	*
Mid Flood	-1.678	0.237	-7.076	<0.001	*

TABLE 24. PERMANOVA ON THE RELATIVE ABUNDANCE OF MAJOR ZOOPLANKTON TAXA IN DAYTIME SAMPLES. ANALYSIS DOES SHOW DIFFERENCES BETWEEN COMMUNITY COMPOSITION IN SURFACE VERSUS BENTHIC SAMPLES, AND A TREND TOWARD A DIFFERENCE IN WETLAND VERSUS CHANNEL SAMPLES.

Term	DF	Sums of Sqs.	Mean Sqs.	f-value	R ²	p-value	
Tide	3	0.108	0.036	3.290	0.191	0.006	*
Wetland/Channel	1	0.033	0.033	3.016	0.058	0.054	.
Surface/Benthic	1	0.097	0.097	8.880	0.172	0.001	*
Residuals	30	0.329	0.011	0.580			
Total	35	0.567	1				

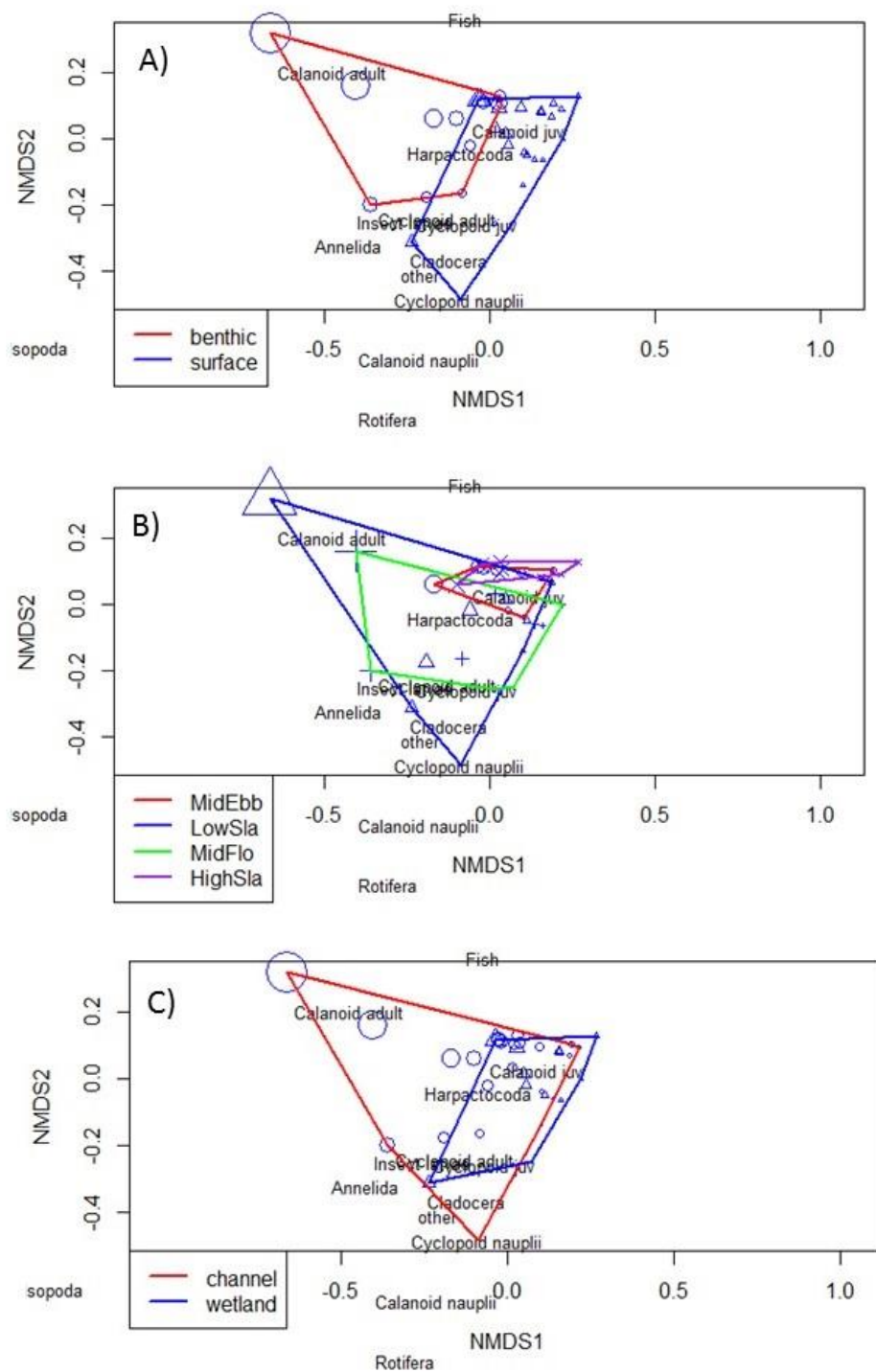


FIGURE 27. NMDS PLOT OF RELATIVE ABUNDANCE OF ZOOPLANKTON IN ALL DAYTIME SAMPLES FROM THE 24 HOUR STUDY (STRESS = 0.032). POINTS REPRESENT SAMPLES, TEXT REPRESENTS SPECIES. SAMPLE POINT SIZE VARIES BY THE PROPORTION OF ADULT CALANOID COPEPODS. A) NMDS PLOT WITH HULLS AROUND BENTHIC AND SURFACE SAMPLES. PERMANOVA SUPPORTS THESE GROUPS BEING SIGNIFICANTLY DIFFERENT. B) NMDS PLOT WITH HULLS AROUND SAMPLES FROM DIFFERENT TIDAL STAGES. PERMANOVA SUPPORTS SIGNIFICANT DIFFERENCES. C) NMDS PLOT WITH HULLS AROUND CHANNEL AND WETLAND SAMPLE. THESE GROUPS HAD A TREND TOWARD BEING DIFFERENT ($p = 0.054$).

Seasonal differences

Macroinvertebrate abundance increased linearly over the course of the spring. AICc supported the linear model over the quadratic model, and R^2 was slightly higher for the linear model (Table 25). Abundance of Delta Smelt adults caught in the SKT survey was highest in January, with declining abundance over the course of the spring (Figure 28). Chinook Smolt abundance at Chipps Island was highest in May, with a bell-shaped distribution over the course of the spring (Figure 28). The highest combined relative abundance of salmon smolts and smelt adults occurred in April, when a large percentage of the macroinvertebrates were also present (Figure 28).

TABLE 25. FIT OF LINEAR MODEL AND QUADRATIC MODEL OF MACROINVERTEBRATE CPUE OVER FIVE SAMPLING PERIODS AT DECKER ISLAND.

Model	DF	f-value	p-value	R^2	AICc	$\Delta AICc$
$\log(CPUE) \sim \text{Date} + \text{geartype}$	68	41.91	<0.0001	0.694	239.04	0
$\log(CPUE) \sim \text{Date}^2 + \text{geartype}$	68	40.15	<0.0001	0.685	241.26	2.22

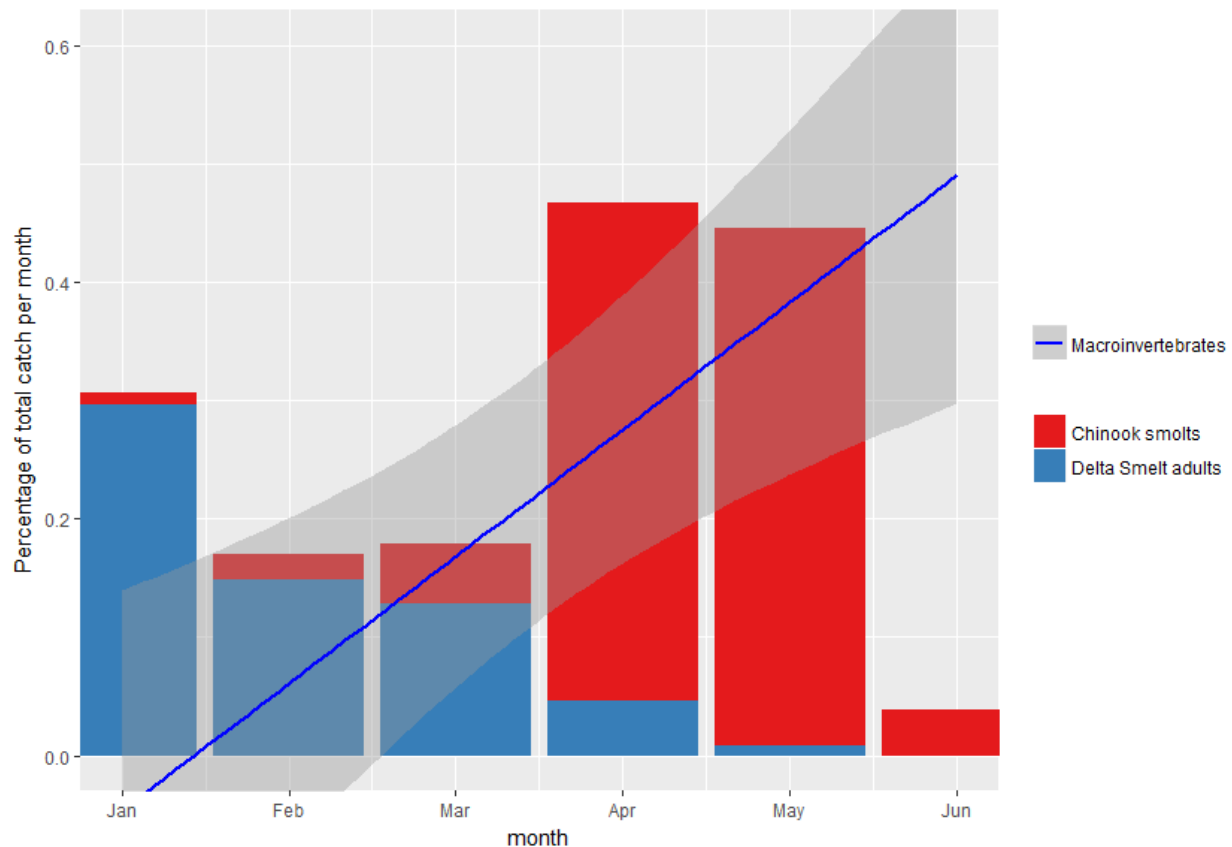


FIGURE 28. PROPORTIONAL CATCH OF DELTA SMELT ADULTS AND CHINOOK SALMON SMOLTS PASSING THROUGH THE DELTA PER MONTH AS MEASURED BY THE SPRING KODIAK TRAWL AND THE CHIPPS ISLAND TRAWL. A LINEAR MODEL OF MACROINVERTEBRATE CATCH WITH 95% CONFIDENCE INTERVAL IS OVERLAID.

Discussion

Tidal and diel differences

Our 24 Hour Study found strong evidence for both tidal and diel migration in calanoid copepods during the summer in the lower Sacramento River. The significant main effect of tide in the model of overall CPUE and the model of calanoid adult CPUE indicates adult calanoid copepods are more abundant during high slack tide. However, the interaction with time of day shows this effect is less pronounced at night, when calanoid copepods are more common in the water column at all tidal stages (Figure 22, Figure 24, Table 19). This is similar to studies by Kimmerer et al. (1998), who found higher abundance of all zooplankton at night than during the day, and a higher abundance on flood tides than ebb tides. This pattern was much more common in copepods than other zooplankton (Kimmerer et al. 2002). Tidal migration by zooplankton is thought to be an important mechanism for maintaining position in the estuary (Kimmerer et al. 2014, and references therein; Orsi 1986), or transporting to more favorable salinities (Manuel and O'Dor 1997), and may also be important in calculating zooplankton export from wetlands (Dean et al. 2005). One important caveat: the high slack samples during our study were taken at dusk and dawn, rather than truly day and night, so our results may not be indicative of overall trends.

With the calanoid copepods removed from the dataset, we still found evidence for diel migration, with higher abundances in the surface water at night (Table 22, Figure 25), but no longer found evidence for

tidal migration. Diel migration is thought to chiefly be a predator avoidance mechanism – sinking during the day to avoid visual predators, and rising at night to graze on phytoplankton (Lampert 1989; Manuel and O'Dor 1997). Diel migration has been found frequently in a wider variety of invertebrates than tidal migration, including copepods, mysids, Cladocera, amphipods, and even chironomid larvae (Kimmerer et al. 1998; Marklund et al. 2001; Rollwagen-Bollens et al. 2006).

With the decrease in abundance of zooplankton, particularly adult calanoid copepods, during the day for most tidal stages, there was a corresponding increase in zooplankton in benthic samples (Figure 27, Figure 26, Table 23), similar to the abundance of zooplankton at night (Figure 22, Figure 26). This suggests zooplankton were sinking to bottom where they could better escape both predation and strong currents. While the wetland samples were taken in relatively shallow water (< 3 m), and so had only a single tow, a future study could try to better target both surface and benthic samples in shallow water adjacent to wetlands.

This study was conducted over a single 24-hour period, during which environmental parameters were relatively homogeneous (data not shown), so we cannot make inferences about what other factors influence vertical migration. However, high turbidity has been shown to decrease incidence of vertical migration (Dodson 1990), and the mysid shrimp *Neomysis mercedis* may or may not exhibit vertical migration, depending on environmental circumstances (Kimmerer et al. 2002; Orsi 1986). The lunar cycle may also influence vertical migration, with a lower incidence of migration on full moons (Manuel and O'Dor 1997). The San Francisco Estuary has mixed semi-diurnal tides, meaning each daily tidal cycle has a “high-high” and a “low-high”. In our study, the low-low tide occurred during the day, and the high-high tide occurred during the night (Figure 21). The magnitude of the change in tide may affect the degree of vertical migration.

We found few differences in zooplankton community composition or abundance in the wetland trawls versus the channel trawls. This is similar to a study by Grimaldo et al (2004), who found some differences in channel versus wetland abundance by species, but no overall differences in zooplankton abundance, including for the dominant taxa, *P. forbesi*. Kimmerer and Slaughter (2016) also studied fine-scale distributions of *P. forbesi*, and did not find significant differences in channel versus shallow-water habitat, but shallow-water samples were not along wetlands. Because our samples were along edges of wetlands in a relatively well-mixed river channel, we may have found greater differences in zooplankton communities deeper into the wetland.

We predicted that shallow-water samples might exhibit less evidence for vertical migration than channel samples, because the water was shallow and largely vegetated. However, the lack of a significant interaction between the Channel/Wetland term and Day/Night term in our model demonstrated no difference in migration patterns (Table 19). Some invertebrates associated with vegetation do exhibit vertical migration even within dense stands of submerged aquatic vegetation (Marklund et al. 2001), and previous studies showing higher larval fish abundance in shallow water (Grimaldo et al. 2004) and vegetation (Young et al. 2018) may mean vertical migration for predator avoidance is even more important in shallow water than the open channels.

Larval Delta Smelt may exhibit some diel vertical migration as well, tracking their zooplankton food through the water column, though few studies have been able to sample with adequate replication to fully explore the issue. One study found higher larval smelt abundances at night than during the day at any depth, with no evidence for tidal migration (Rockriver 2004), while another study found greater

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abundances at the surface during the day, though there is potential for ontogenetic shifts in migration patterns (Bennett et al. 2002). Longfin Smelt larvae also exhibited tidal migration (Bennett et al. 2002), which may help fish maintain their position in the estuary as well as track their zooplankton food supply. Adult smelt also move in response to tides, moving into shallow-water embayments on high tides (Aasen 1999), and moving to channel edges during ebb tides during high flow events (Bennett and Burau 2015). The interplay of fish movements with invertebrate migration may impact when productivity is most available for the fish of interest to eat.

Moving forward in monitoring tidal wetland restoration sites, this study demonstrates that tidal and diel migration may greatly influence export rates of certain zooplankton taxa, especially *P. forbesi*. While we may not be able to sample night and day on a regular basis, knowing that daytime abundances of copepods are an underestimate will help in making inferences about the community as a whole.

Seasonal differences

Sampling macroinvertebrates at Decker Island with high temporal frequency demonstrated that abundance of most macroinvertebrates increases linearly over the course of the spring (Jan-Jun; Table 25), most likely peaking sometime in the summer after sampling had ended. Salmon smolts and adult smelt both had peak abundance significantly before the peak in macroinvertebrate abundance, with adult smelt being most abundant in January and salmon being most abundance in May. However, by choosing to sample intensively for macroinvertebrates in April, we can target a time period when macroinvertebrates are increasing in abundance, adult Delta Smelt are still present, and salmon smolts are near their maximum abundance (Figure 28).

The macroinvertebrate data used for this analysis is from a single year of sampling, whereas the fish data were summarized from several years of surveys. Because 2017 was a very wet year (Figure 29), it is possible that other water year types may not show the same trend. Many studies have found differences in invertebrate abundance year-to-year, which is generally attributed to differences in water flow (Bollens et al. 2014; Crauder et al. 2016; Hennessy and Enderlein 2013; O'Rear and Moyle 2013). However, multiple other studies of macroinvertebrate and zooplankton abundance show peaks during the summer in many different water year types (Corline et al. 2017; Hennessy and Enderlein 2013; Howe et al. 2014). In 2018, FRP repeated the temporally intensive sampling at Decker Island (sample processing ongoing as of September, 2018), and results of this second year of sampling may shed light on inter-annual differences in seasonal macroinvertebrate trends.

This study was conducted at Decker Island, which is one site in a dynamic estuary. Community composition of both fish and macroinvertebrates is highly variable across the estuary (Bollens et al. 2014; Feyrer et al. 2017; Thompson et al. 2013), and while overall trends in abundance tend to be similar, there may be some restoration sites with peak fish abundances later than others. In particular, Delta Smelt migrating from fresh water to the low salinity zone may be taking advantage of macroinvertebrates in Suisun later in the year than those in the freshwater Delta (Baxter et al. 2015). In 2018, FRP will begin a second round of macroinvertebrate sampling in the fall nearby sites where smelt may be located later in the year.

We do not present a detailed analysis of change in invertebrate community composition here, but we did not detect differences in relative abundance in certain taxonomic groups between sampling periods. In choosing a single spring sampling point for spatially intensive invertebrate sampling, we wanted to

evaluate the total invertebrate abundance, but if a special study were to target a particular taxon, they may find different results. For example, the timing of peak of abundance is different for different species of mysids and varies by region of the estuary (Hennessy et. al. in prep). Howe et al. (2014), also found variation in timing of a broad suite of macroinvertebrates between species and between regions of the estuary. Studies in other estuaries targeting food resources for juvenile salmon frequently find seasonal and inter-annual differences in abundance of different groups of invertebrates (Woo et al. 2017).

Part 3. Sample size and variability of food web data

Introduction

The food web supporting at-risk fishes is based on a foundation of phytoplankton. It builds with scaffolding of aquatic vegetation, epiphytic and epibenthic invertebrates, and pelagic invertebrates. These groups are supplemented by drift invertebrates falling onto the surface of the water from the surrounding upland zone. While all these components contribute to the food supply for salmon and smelt, they do not all contribute equally, and FRP cannot afford to spend undue resources on food web components that are less important, or are too variable to demonstrate effectiveness of a restoration site. Because many of these communities are under-studied, the variability in abundance and community composition of these groups is unclear.

Understanding variability of many food web components will allow us to evaluate appropriate timing and replication of samples, and help focus monitoring efforts on the most efficient metrics of food web support. To evaluate sample size, we conducted a single, large spatially intensive sampling effort of zooplankton, benthic macroinvertebrates, epiphytic invertebrates, neuston (surface) invertebrates, chlorophyll-a, and phytoplankton at sites distributed across the Delta and Suisun Marsh.

Both mesozooplankton (those retained by the 150 μm mesh net), and larger open-water invertebrates (retained by a 500 μm mesh net) are regularly sampled in channels throughout the estuary. While previous research has demonstrated zooplankton communities to be distributed fairly even across major channels (Kimmerer and Slaughter 2016), it is unclear how well-distributed zooplankton communities may be in wetlands. Wetlands often have dendritic channels, ponds, and pannes that may have a variety of water quality, connectivity, and habitat attributes that may make zooplankton more variable within wetlands than in open-water habitats (as found by Cooper et al. 2012), as has been seen for fish community composition (Williams and Zedler 1999).

Benthic invertebrates are not commonly consumed by smelt, however, epibenthic amphipods and chironomids are major components of salmonid diets (David et al. 2014). Many of these taxa also have pelagic life stages where they are more available to both salmon and smelt. Furthermore, benthic filter-feeders, particularly invasive clams, may compete with zooplankton for phytoplankton food resources, or directly consume early life stages of zooplankton (Kimmerer and Lougee 2015). Benthic invertebrates are sampled by DWR's Environmental Monitoring Program, but benthic invertebrate distributions are extremely patchy (Howe et al. 2014; Peterson and Vayssières 2010), so we cannot expect channel samples to reflect clam abundance inside wetlands.

Macroinvertebrates associated with vegetation and shallow water habitat, such as amphipods and insect larvae, provide the majority of salmonid diet composition in estuarine habitats (Bottom et al. 2011; David et al. 2016; Maier and Simenstad 2009; Sommer et al. 2001). They are also a component of Delta Smelt diets when smelt occur in areas of high macrophyte production (Whitley and Bollens 2014). However, epiphytic invertebrates are extremely patchy and difficult to sample in a standardized way (Contreras et al. 2017; Contreras et al. 2016), so are still under-studied in this system.

Surface invertebrates, including emerging insects and fall-out invertebrates, are also poorly studied in the Estuary, but are a key food resource for salmonids. We can make few inferences on the variability of

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these communities, but data from the Yolo Bypass Fish Monitoring Program shows that surface invertebrates are much more abundant in the Toe Drain, adjacent to wetland habitat, than the mainstem Sacramento River (Jared Frantzich, DWR, pers. Comm.). Other special studies of these communities indicate they may be highly variable within wetland habitat (Howe et al. 2014; Simenstad et al. 2013).

Phytoplankton abundance and community composition may be critical to understanding wetland export of food resources. While detrital carbon may be more important in wetlands than in pelagic food webs, phytoplankton is still the most available form of carbon for zooplankton (Sobczak et al. 2002). However, not all phytoplankton is created equal. Centric diatoms are considered “high value” due to their large size and high essential fatty acid content, whereas cyanobacteria are small and less nutritious (Galloway and Winder 2015). In some cases, cyanobacteria may be toxic to zooplankton (Ger et al. 2010). Ensuring our restored wetlands produce the right kind of phytoplankton will be key to understanding the effectiveness of our actions.

Study questions:

1. What is the variability in phytoplankton, zooplankton, and macroinvertebrates in and near wetland restoration sites? This will help us determine sampling design for long-term monitoring. Specifically:
 - a. What is the variability between habitats within a site (island or defined reach of channel), and is it greater or less than variability between sites?
2. Are there significant differences between channel habitat, managed wetlands (pre-restoration), and tidal wetlands (reference and/or post-restoration)?
3. How many samples are necessary to adequately answer questions 1 and 2?

Sampling sites

We distributed sites across the Delta and Suisun Marsh to incorporate varying salinity and surrounding land use (the “ecoclines” identified in the IEP TWM PWT conceptual models, see Hartman and Sherman (2017)). We grouped these sites into four regions that include existing managed and tidal wetlands and adjacent open water or channels (Figure 30, Table 26). This information on broad-scale spatial variability will also be useful for baseline abundance estimates in later evaluations of restoration projects that are planned for these regions. We sampled most sites during a single spring sampling event in late March and early April, 2017 (4-10 samples per habitat type, depending on wetland size, see Table 26). This was after the peak high flows in February (Figure 29), when water was starting to warm and productivity generally increases. This is also the period when juvenile salmon are rearing in the Delta (del Rosario et al. 2013), and most Delta Smelt adults have already migrated to spawn, so are able to make use of wetland resources through a larger extent of the estuary (Baxter et al. 2015).

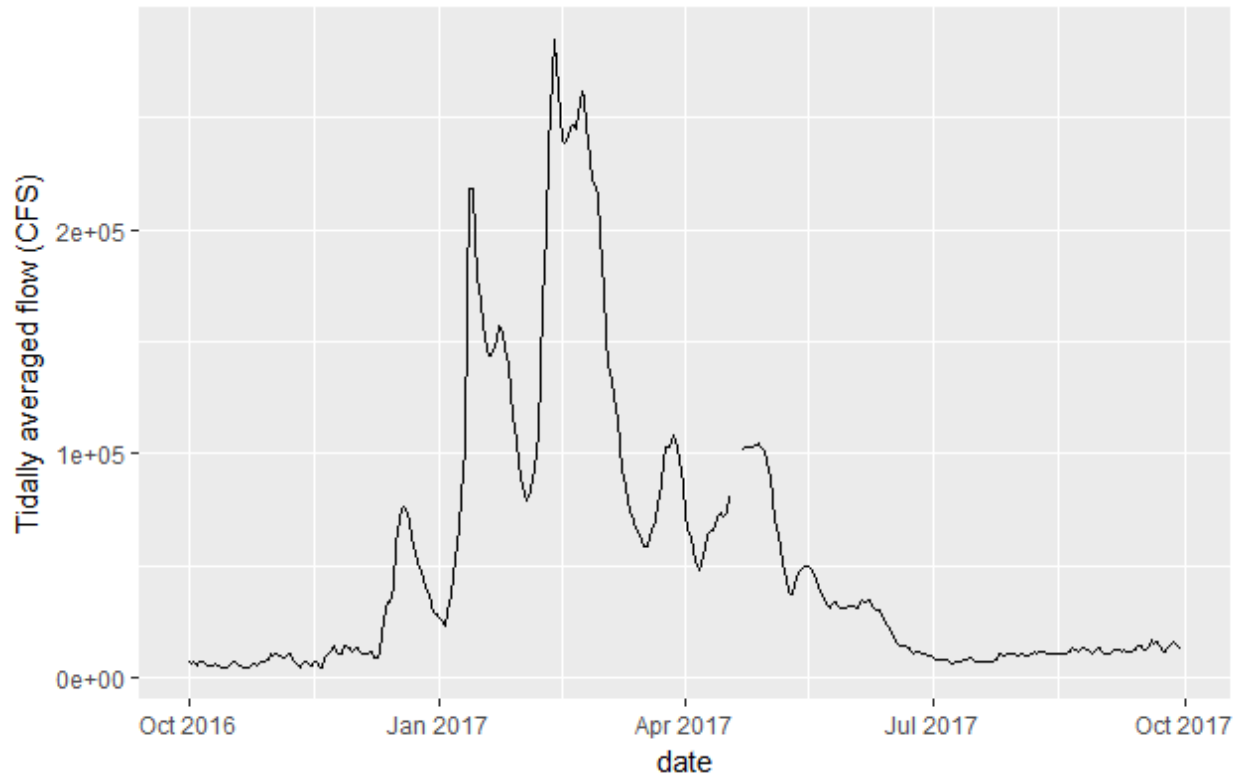


FIGURE 29. TIDALLY AVERAGED FLOW IN CUBIC-FEET PER SECOND (CFS) IN THE SACRAMENTO RIVER AT RIO VISTA (USGS STATION #11455420). DATA FROM:
[HTTPS://WATERDATA.USGS.GOV/NWIS/INVENTORY?AGENCY_CODE=USGS&SITE_NO=11455420](https://waterdata.usgs.gov/nwis/inventory?agency_code=USGS&site_no=11455420).

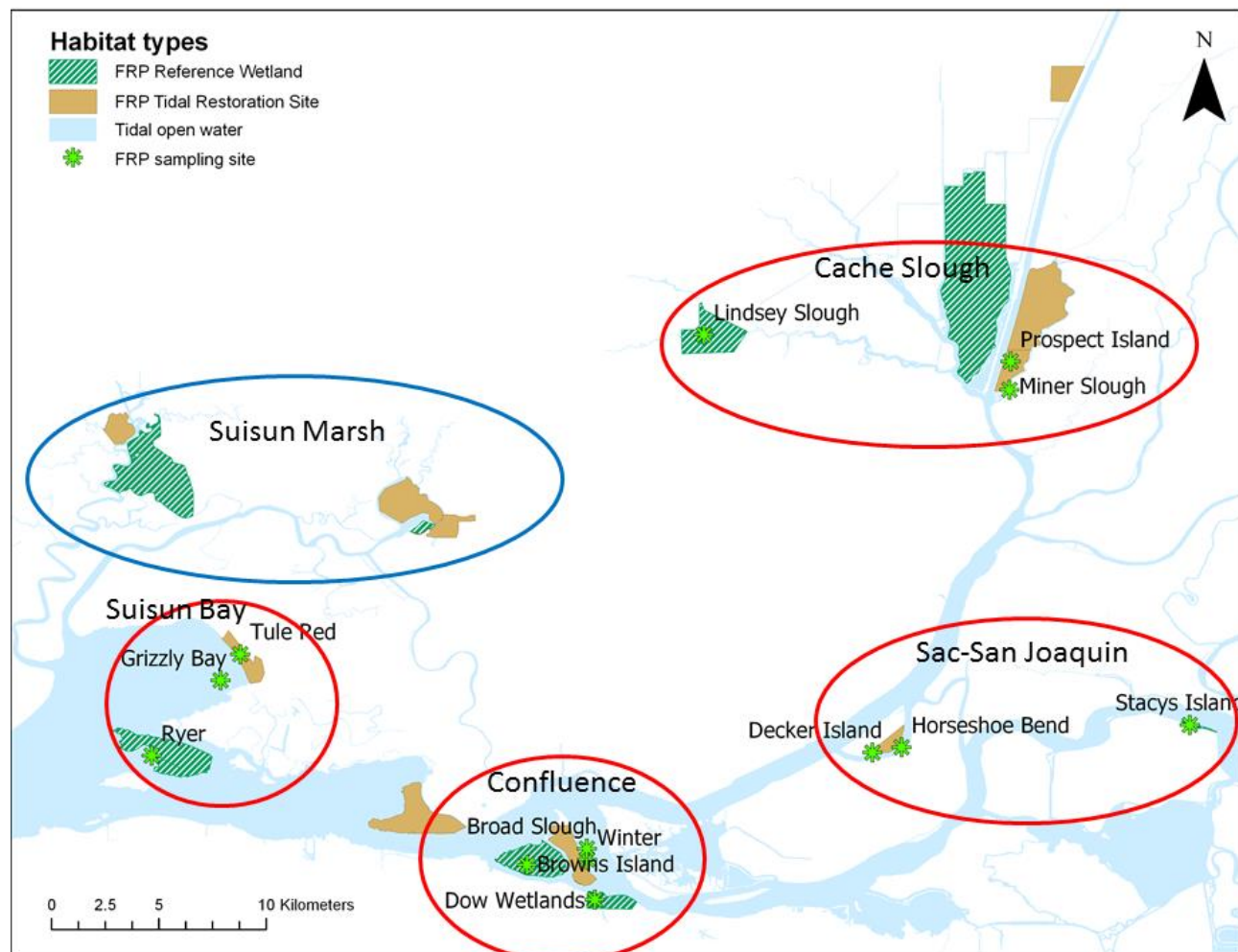


FIGURE 30. SITES THAT WERE SAMPLED IN PHASE III. WITHIN EACH REGION (OUTLINED IN ORANGE), WE COMPARE PRE-PROJECT DATA AT PLANNED RESTORATION SITES (WHERE RELEVANT) AND FROM CURRENTLY MANAGED OR MUTED TIDAL WETLANDS, WITH DATA FROM ASSOCIATED CHANNEL HABITAT AND POST-RESTORATION DATA (WHERE AVAILABLE). THE NURSE SLOUGH COMPLEX WAS SAMPLED FOR FISH AND ZOOPLANKTON, BUT NOT FOR MACROINVERTEBRATE MONITORING IN 2017 DUE TO LOGISTICAL LIMITATIONS.

TABLE 26. SAMPLING SITES AND SAMPLE SIZES FOR THE SPRING SAMPLING BOUT, MARCH-APRIL 2017.

Site	Region	Site type	Phytoplankton	Chlorophyll	EAV	SAV	FAV	Benthic core	Mysid trawl	Neuston Trawl	Zoop trawl
Winter Island	Confluence	muted tidal	5	7	9	0	4	3	4	4	4
Dow Wetlands	Confluence	muted tidal	4	6	8	0	6	0	5	5	5
Browns Island	Confluence	tidal wetland	3	5	5	6	1	5	6	6	6
Broad Slough	Confluence	channel	3	4	6	0	0	2	5	5	5

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Tule Red	Grizzly Bay	managed	6	6	5	3	0	5	0	0	5
Ryer Island	Grizzly Bay	tidal wetland	3	3	5	3	0	3	3	3	3
Grizzly Bay	Grizzly Bay	channel	2	2	0	0	0	3	4	3	4
Decker Island	Sac River	muted tidal	3	3	7	0	3	1	11	7	10
Stacy's Island	Sac River	tidal wetland	3	3	6	0	3	2	2	3	3
Horseshoe Bend	Sac River	channel	4	5	5	6	6	6	6	6	6
Prospect Island	Cache Slough	muted tidal	6	7	7	1	7	0	5	6	10
Miner Slough	Cache Slough	channel	0	0	3	0	3	0	5	0	4
Lindsey Slough	Cache Slough	tidal wetland	6	8	6	5	6	7	6	6	6
Total			48	59	72	24	39	37	62	54	71

Habitat types and sampling gears

Habitat type (water depth and presence of vegetation) impacted efficacy of our sampling methods, so we tested methods in four different habitat types (Figure 31). Not all methods could be applied in all habitats.

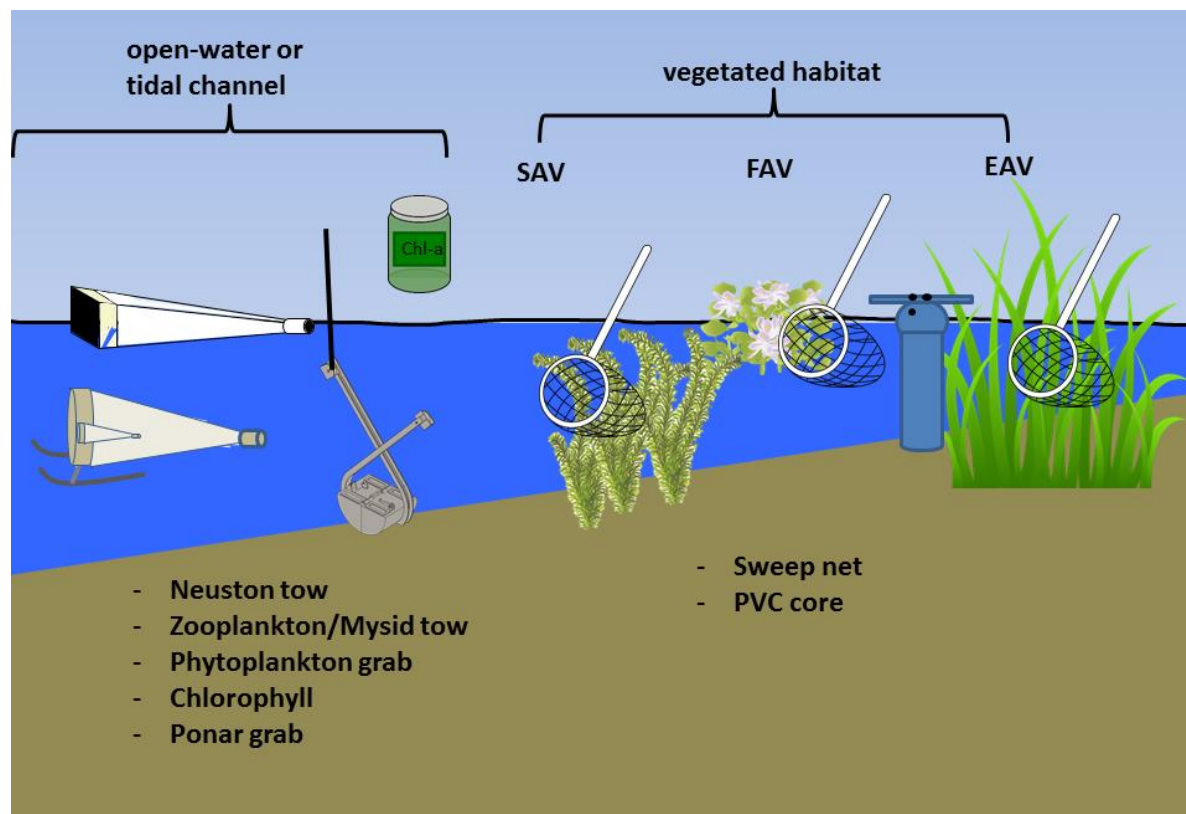


FIGURE 31. HABITAT TYPES AND RELEVANT GEAR TYPES USING IN SPRING OF 2017. NOT ALL SAMPLING TYPES WERE RELEVANT AT ALL SITES.

Vegetated habitats: We used a 25 cm x 30 cm d-frame net with 500 μ m mesh for all sweep net samples. We haphazardly choose 4-10 sampling locations per site and conducted sweep nets in vegetation types roughly proportional to their coverage of the site. We adapted the sweep net technique slightly for different habitat types.

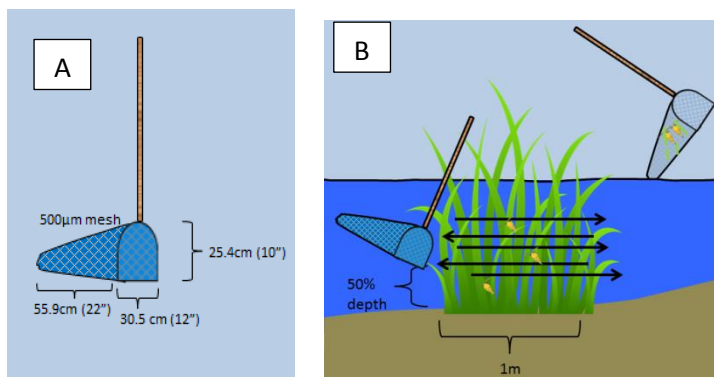
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Emergent Aquatic Vegetation (EAV): Five 1 m sweeps, scraping the vegetation as much as possible to knock invertebrates off the stems. All samples were taken within 1 m of the edge, since vegetation was less dense and easier to sample. This is also the area where salmonids have been shown to forage most effectively (Simenstad and Cordell 2000). Samples were taken in whichever species was dominant at the site, usually *Schoenoplectus acutus*, *Schoenoplectus californicus*, or *Phragmites australis*.

Submerged Aquatic Vegetation (SAV): Five 1 m sweeps through the thickest growth, collecting vegetation remaining within the frame of the net at the end of the sweep. In the laboratory, vegetation was dried to a constant weight. Samples were taken in whichever species of vegetation was dominant, most frequently *Egeria densa*, with some samples in *Ceratophyllum demersum* and *Stuckenia pectinata*.

Floating Aquatic Vegetation (FAV): The net was lifted from beneath a clump of FAV. Plant material outside of the net frame, and any leaves above the surface of the water, were severed from the sample with shears (similar to Donley Marineau et al. 2017). In the laboratory, the roots were dried to a constant weight. Samples were taken in whichever species of FAV was dominant, most frequently *Eichhornia crassipes*, with some samples in *Ludwigia* spp. and *Azolla* spp.

EAV sample catch per unit efforts (CPUE) were calculated as number of invertebrates per sample (five sweeps). CPUEs for SAV and FAV samples were calculated as number of invertebrates per gram of dried vegetation.



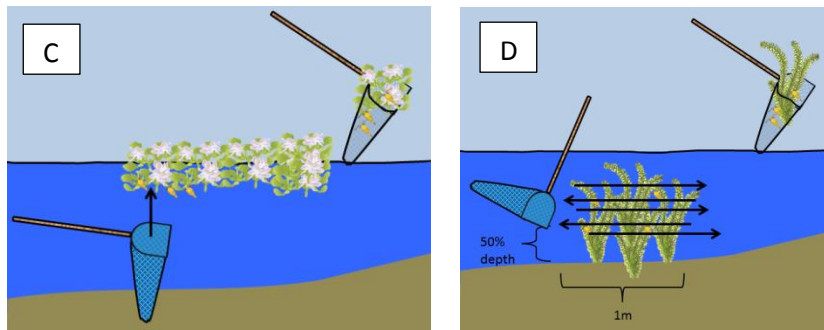


FIGURE 32. A) SPECIFICATIONS OF THE SWEEP NET. B) USE OF SWEEP NET IN EMERGENT VEGETATION. C) USE OF SWEEP NET IN FLOATING VEGETATION. D) USE OF SWEEP NET IN SUBMERGED VEGETATION.

Open water and channel:

Our open water sampling patches were haphazardly distributed across all unvegetated open water and channels > 1.5 m across. We combined open-water sampling with fish sampling where possible to reduce disturbance and increase efficiency. Methods used in open-water have a long history of use in monitoring in the Delta, and allowed us to compare our measurements in vegetated wetlands to conditions in channels and make comparisons to long-term data sets. Methods included: zooplankton tow, mysid (macrozooplankton) tow, neuston tow, and benthic cores/ponar grabs.

Zooplankton and Mysid tows: Zooplankton and mysid tows used the same methods described above in Part 1, Gear Descriptions. Paired zooplankton and mysid tows were distributed across wetland channels and towed for five minutes, or held in the current at the mouth of a channel on the ebb tide.

Benthic core: Benthic cores have been used extensively to quantify chironomid and amphipod populations, as well as bivalves and other infauna in tidal wetlands (Howe et al. 2014; Wells 2015). We had some difficulty predicting appropriate tidal stage for sampling at some of the sites, so sample size was small and unbalanced. Sampling during the 2018 work plan was more evenly distributed.

In shallow water (< 1.5 m), we took a 4 in (20 cm) diameter benthic core (Figure 33A), hand-deployed to a depth of 20 cm. In deep water > 1.5 m, we used a 9 x 9 in ponar grab modified for use in hard substrates (as per USFWS Liberty Island Monitoring, L. Smith pers. comm, Figure 33B), with three samples at each site. The core was washed and sieved on board the boat to remove the sand/mud and preserve any organic detritus and invertebrates. Two crew members estimated % silt, sand, and gravel in the field, and averaged the values. Effort was calculated as catch per surface area of substrate sampled.

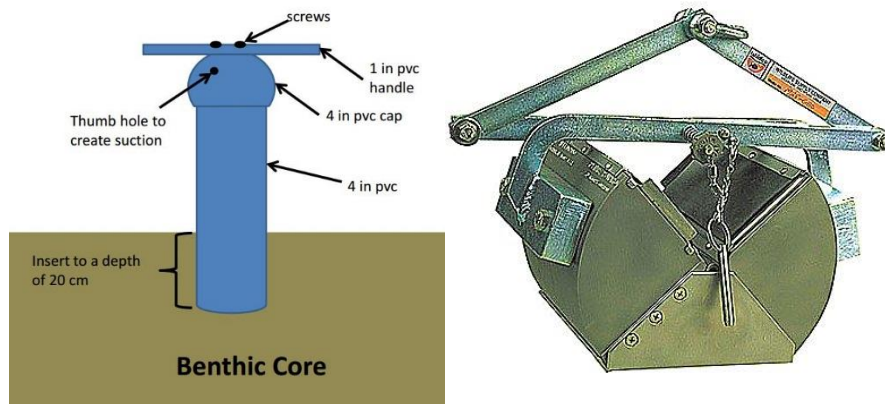


FIGURE 33. A) BENTHIC CORE MADE OF 4 IN PVC PIPE FOR USE IN SHALLOW WATER (< 1.5 METERS). B) PONAR GRAB FOR USE IN WATER GREATER THAN 1.5 METERS.

Neuston tow: Emerging insects and Collembola found at the surface of the water are an important feature in salmonid diets, and are commonly sampled using neuston tows and drift nets (Sommer et al. 2001, Howe et al. 2014). The neuston net is a 45 cm x 30 cm rectangular net, 1 m long with 500 μ m mesh towed half-way out of the water to sample invertebrates on the surface of the water (Figure 32). We towed the neuston net at the surface of the water from the side of the boat via a boat-hook. In narrow channels, we pulled the net along the edge of emergent vegetation by hand (as in Howe et al. 2014). We standardized effort by the distance of the tow calculated by GPS track multiplied by width of net to calculate surface area of water sampled. After retrieval, all content collected in a cod end was preserved in 70% ethanol for later ID.

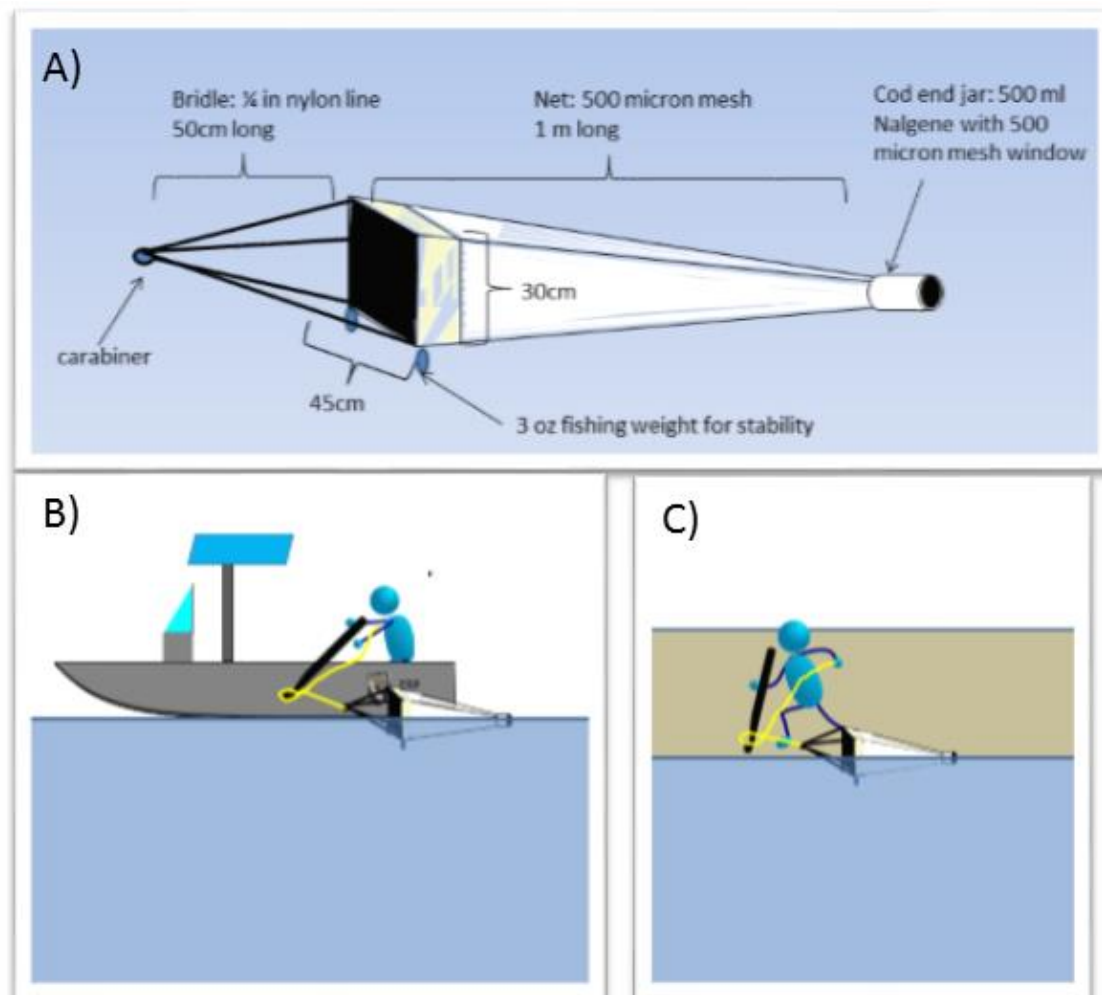


FIGURE 34. A) SPECIFICATIONS FOR THE NEUSTON NET. B) DEPLOYING THE NEUSTON NET ALONGSIDE A BOAT. C) DEPLOYING THE NEUSTON NET FROM SHORE.

Phytoplankton: We sampled phytoplankton community composition and chlorophyll-*a* concentration concurrently with our spring macroinvertebrate sampling in order to best characterize differences in site type and region of the estuary. We collected between three and six samples haphazardly distributed across each site, following the PWT Water Quality Grab Sample SOP (PWT 2017b). All phytoplankton samples were preserved in Lugol's iodine solution and transported to a contracting lab (EcoAnalysts, Inc, Moscow, Idaho) for enumeration. All chlorophyll samples were filtered, frozen on dry ice, and transported to DWR's Bryte Laboratory for analysis.

Laboratory methods

Invertebrate and zooplankton samples: Laboratory methods for all invertebrate samples followed the same methods in Part 1. Laboratory methods, above.

Mesozooplankton (Copepoda, Cladocera, and Rotifera) occurring incidentally in macroinvertebrate samples (mysid nets, neuston nets, benthic samples and sweep nets) were enumerated during sorting, but data on these taxa were removed during analysis of the whole macroinvertebrate data set because they are more accurately quantified using the zooplankton net. After analyzing the macroinvertebrate

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data set as a whole, we ran a separate analysis of sweep net samples in different types of vegetation with both macroinvertebrates and mesozooplankton included, since zooplankton nets could not be used effectively in vegetated habitat.

Phytoplankton: All chlorophyll samples were analyzed by Bryte Labs for chlorophyll-*a* and pheophytin-*a* using Standard Method 10200 H (APHA 2017). All community composition samples were analyzed by EcoAnalysts, Inc. (Moscow, ID), using the Utermöhl microscopic method (Utermöhl 1958) and APHA Standard Methods (APHA 2017). In brief: At least 400 total algal units and 100 units of the dominant taxon or taxa (genus or species level) were counted at appropriate levels of magnification for the cell size. The count of the major taxon counted towards the total units, which was at least 400 total. Final counts were expanded to account for subsampling.

Analysis

We first calculated catch-per-unit-effort (CPUE) for each sample using formulas listed for the relevant gear type, above. To answer Questions 1 and 2 on the variability and appropriate sample size between sites, we compared CPUE and community composition of samples from our spatially extensive spring sampling event. We used generalized linear mixed models (GLMM) to compare total abundance across habitat types within sites, among sites of different management types, and among regions of the estuary as listed in Table 27 (Gotelli and Ellison 2012). We included Site as an error term because some regions had multiple sites within a site type. Mixed models were run using R package “lme4” and “lmerTest” (Bates et al. 2016; Kuznetsova et al. 2017). Separate models were run on macroinvertebrate samples, mesozooplankton samples, and chlorophyll-*a* samples. Data were log-transformed where necessary to meet assumptions of the model.

When modeling community composition, we calculated the relative percent composition of each taxon in each sample to separate changes in community composition from changes in abundance. We performed a PerMANOVA on the matrix of relative percent composition (R package “vegan” function “adonis”, Oksanen, 2016) using Region, Site type, and Habitat type as predictors (see Table 27). To examine results visually, we ran an NMDS on the same matrix and plotted it with hulls around groups for each predictor variable, followed by hypothesis testing of NMDS hull centroids (R Package “vegan” functions “monoMDS” and “envfit”) (Oksanen et al. 2016). Separate multivariate analyses were run on phytoplankton data, mesozooplankton data, and for each sampling type individually for the macroinvertebrate data.

TABLE 27. PREDICTOR VARIABLES USED IN GLMMs OF LOG-TRANSFORMED CPUE FOR MACROINVERTEBRATES.

Variable	Variable type	Description	Interpretation
Region	Categorical	Region of the estuary as shown on Figure 30	Higher invertebrate abundance in certain regions
Site type	Categorical	Depth and water management regime (Managed wetland, tidal wetland, muted tidal wetland, or channel)	Different invertebrate abundance in wetlands and channels

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Gear Type	Categorical	Major type of invertebrate community targeted (Vegetation [EAV, SAV and FAV], mysids, neuston, or benthic). Vegetation was lumped together for main analysis, then separated into sub-types for sweep-net analysis.	Used as a blocking variable, different gear types have different CPUE calculations.
Site	Categorical	Specific wetland sample was collected from.	Included as a random term to avoid pseudoreplication.

To answer Question 3, we conducted post-hoc power analyses on linear mixed models of log-transformed CPUE to see how many replicates are necessary to differentiate between regions within the estuary and between site types within a region using the R package “simr” (Peter and J. 2016). This program uses Monte Carlo simulations to calculate power from generalized mixed models, and artificially expands the data set by replicating values to extrapolate power for greater sample sizes. We also calculated the coefficient of variation for each habitat type within a given site, and compared mean within-site variation to the coefficient of variation of group means between sites.

Results

When analyzing the entire dataset of all macroinvertebrate data, there was significantly different CPUE in different site types within a region, but no significant difference between regions. Specifically, there was significantly higher catch in managed wetlands than any other site type, and higher catch in tidal wetlands than channel habitat. There were significant differences in habitat types within a site, but because they were sampled with different gear types, they should not be compared directly and are included only as blocking terms (Table 28, Figure 35, Figure 36).

When analyzing community composition of all macroinvertebrate samples combined, the PerMANOVA indicated there were significant differences in relative percent composition of major taxonomic groups between regions, habitat types, and site types (Table 29, Figure 37). However, an NMDS on the entire macroinvertebrate data failed to converge after 1999 permutations, indicating it is difficult to find structure amongst such disparate data sets. Therefore, we divided the data by habitat type in order to display the differences in region and site type more clearly. The mysid and neuston data both showed significant differences between regions, and between site types within a region (Figure 39, Figure 40, Table 31, Table 32). PerMANOVA showed differences in benthic communities between regions, but not between site types (Table 30), whereas the NMDS hulls have significantly different centroids for both region and site type (Figure 38). The number of benthic sample replicates was particularly low and the samples we were able to collect ended up being unbalanced; patterns may become more clear with higher replication.

Sweep net data, with mesozooplankton included, were separated by vegetation type (SAV, FAV, and EAV). We found a significant difference in community composition between vegetation type, site type, and region of the estuary (Figure 42, Table 33). The NMDS hulls had a large amount of overlap, however

they did have significantly different centroids (Figure 42). However, because SAV and FAV samples were extremely unbalanced, and biased toward certain regions of the estuary (FAV does not occur in Suisun Marsh), this analysis might not hold up over the long term.

When analyzing the zooplankton data, there was a significant difference in CPUE between regions of the estuary, with lower catch in the confluence than the other regions. However, there was no significant difference between site types (Table 34, Figure 43, Figure 44). The zooplankton community composition analysis was highly skewed by one site (Tule Red), as can be seen by the much larger, separated hull for “managed wetland” in the NMDS plot (Figure 46). This was driven by the dominance of harpacticoid copepods, ostracods, and “other” (at Tule Red the “other” category was mostly snails, nematodes, and aphid larvae). Therefore, we performed the multivariate analysis with and without this site. There was a significant difference between site type and region, whether or not Tule Red was included (Figure 45, Figure 46, Figure 47, Table 35).

Phytoplankton biomass was assessed using chlorophyll-*a* concentration as a proxy. Chlorophyll-*a* was significantly higher at Tule Red (the managed wetland) than any other site, but there were no other significant differences (Table 36, Figure 48). Phytoplankton community composition analysis also showed highly significant differences between communities from different regions of the estuary and different site types (Figure 49, Table 37), as can be seen by lack of overlap in NMDS hulls (Figure 50).

Power analyses for each data set indicated some communities would need much higher sampling effort than others in order to differentiate in abundance between groups, and this varies by grouping (Table 39). Some sampling types could not differentiate between either site type or region, even given 20 samples per site. However, the combined macroinvertebrate data set could differentiate between site type and region, even when individual sampling types could not. Furthermore, the unbalanced sampling in some of these habitat types in 2017 may be skewing this analysis. Zooplankton samples had the highest power for differentiating between both site type and region. Vegetation samples had the highest within-site coefficient of variation, and vegetation samples (combined) had a higher within-site CV than between site CV (Table 38). While we did find differences between site types and regions in community composition, the high coefficient of variation of CPUE in vegetation samples may mean it will be difficult to make accurate statements about invertebrate productivity.

TABLE 28. FIXED EFFECTS FOR GLMM OF LOG-TRANSFORMED TOTAL CPUE OF ALL SPRING MACROINVERTEBRATE SAMPLES. THERE WERE NO SIGNIFICANT DIFFERENCES IN ABUNDANCE BETWEEN REGIONS OF THE ESTUARY, BUT THERE WAS SIGNIFICANTLY HIGHER CATCH IN MANAGED AND TIDAL WETLANDS THAN IN CHANNELS, AND SIGNIFICANTLY HIGHER CATCH IN MANAGED WETLANDS THAN MUTED TIDAL OR TIDAL WETLANDS. SITE WAS INCLUDED AS A RANDOM EFFECT TO AVOID PSEUDOREPLICATION.

Term	Estimate	SE	DF	t-value	p-value	
Intercept: Cache, Channel, Benthic	7.544	0.470	13.244	16.043	< 0.0001	*
Gear type: Mysids	-7.249	0.315	270.923	-23.011	< 0.0001	*
Gear type: neuston	-8.531	0.333	269.916	-25.598	< 0.0001	*
Gear type: Vegetation	-3.981	0.286	271.895	-13.913	< 0.0001	*

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Region: Confluence	-0.139	0.401	5.721	-0.348	0.740	
Region: Sac-San Joaquin	0.028	0.448	6.971	0.063	0.952	
Region: Suisun	-0.564	0.538	8.058	-1.048	0.325	
SiteType: managed	4.520	0.722	8.367	6.266	0.0002	*
SiteType: muted	0.732	0.436	7.286	1.677	0.136	
SiteType: tidal	1.082	0.370	7.051	2.925	0.022	*

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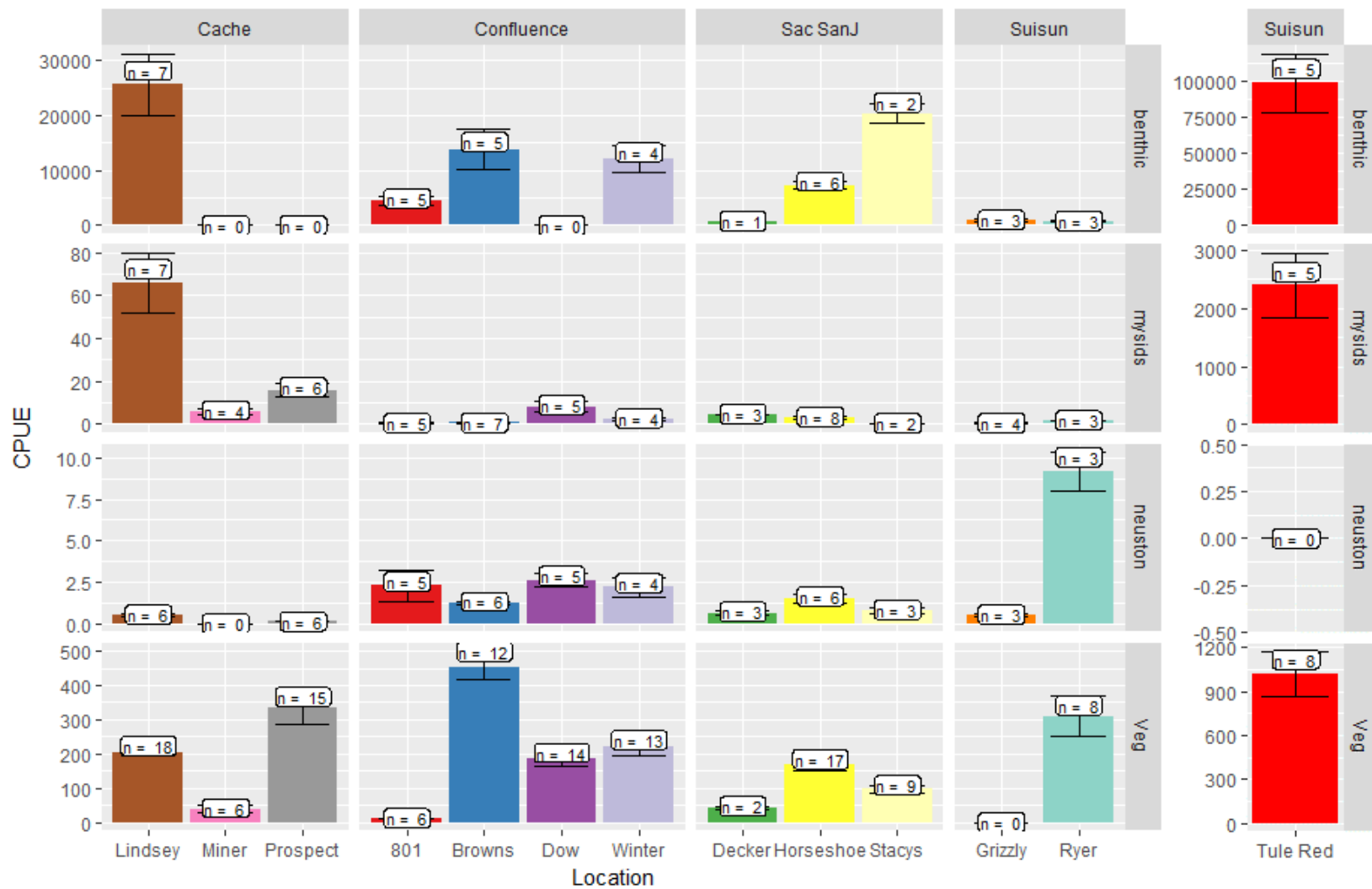
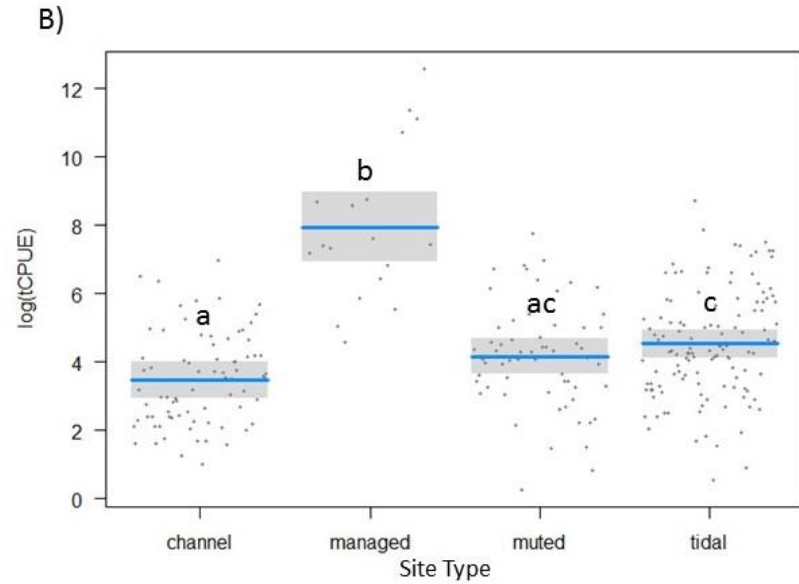
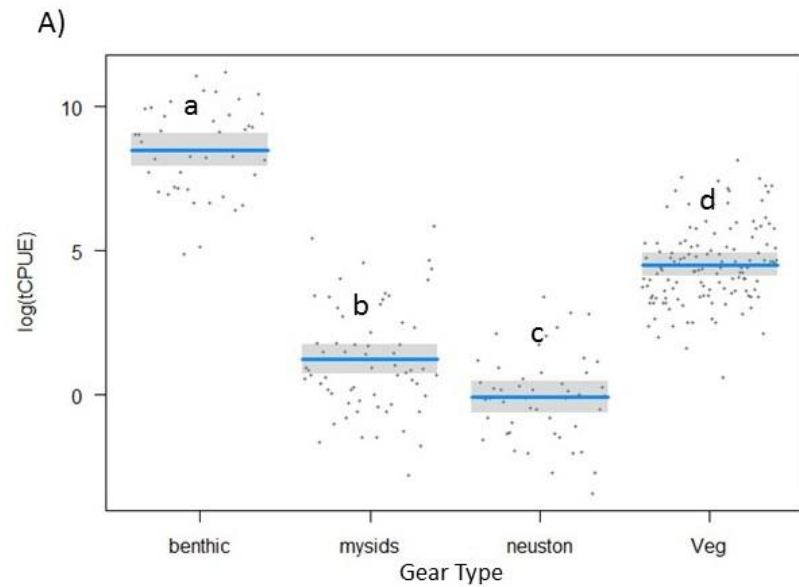


FIGURE 35. MEAN CPUE FOR EACH GEAR TYPE AT EACH SITE, +/- 1 SEM, WITH SAMPLE SIZE LISTED ABOVE EACH BAR. BECAUSE CPUE AT TULE RED MUCH HIGHER THAN ANY OTHER SITES FOR MANY HABITAT TYPES, IT HAS BEEN SHOWN ON ITS OWN AXIS.

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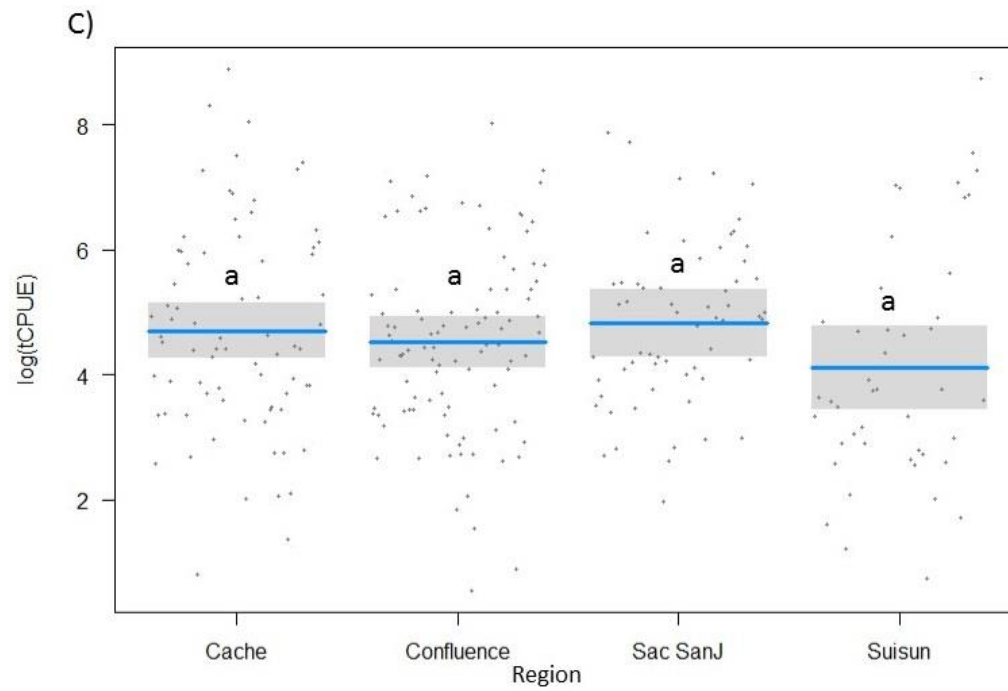


FIGURE 36. PARTIAL RESIDUAL PLOTS FOR EACH TERM IN THE MODEL OF LOG-TRANSFORMED CPUE ON ALL MACROINVERTEBRATE SAMPLES PRESENTED IN TABLE 28. A) GEAR TYPE, B) SITE TYPE, AND C) REGION OF THE ESTUARY. LOWER CASE LETTERS INDICATE GROUPS THAT ARE NOT SIGNIFICANTLY DIFFERENT.

TABLE 29. PERMANOVA ON ALL MACROINVERTEBRATE SAMPLES. 999 FREE PERMUTATIONS, TERMS ADDED SEQUENTIALLY. REGION OF THE ESTUARY, SITE TYPE, AND HABITAT TYPE ALL HAD HIGHLY SIGNIFICANT EFFECTS ON COMMUNITY COMPOSITION. SITE (USED AS A BLOCKING TERM) ALSO HAD A HIGHLY SIGNIFICANT EFFECT ON COMMUNITY COMPOSITION.

Term	Df	Sums of Sqs.	Mean sqs.	f-value	R²	p-value	
Region	3	5.896	1.965	14.139	0.093	0.001	*
Site Type	3	4.529	1.510	10.860	0.071	0.001	*
Gear Type	3	12.188	4.063	29.228	0.192	0.001	*
Site	6	4.008	0.668	4.806	0.063	0.001	*
Residuals	266	36.973	0.139	0.581			
Total	281	63.594	1				

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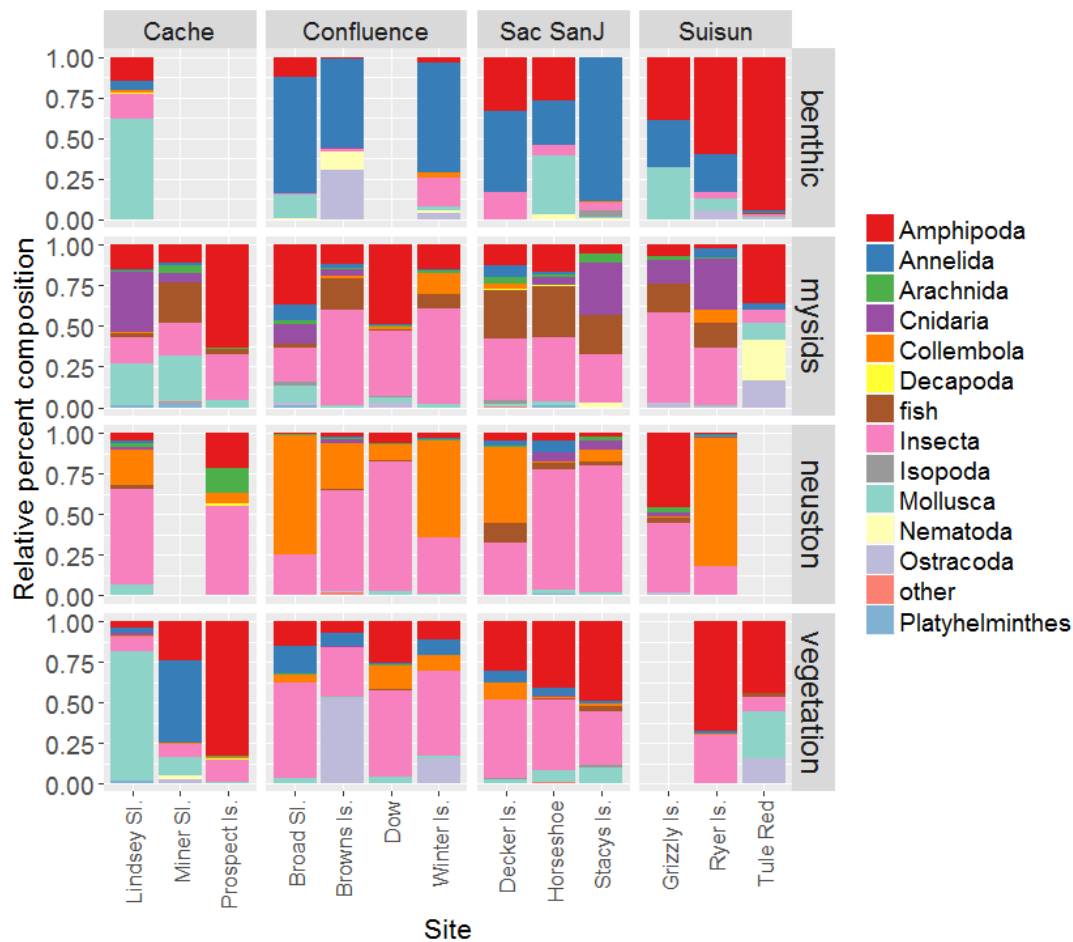
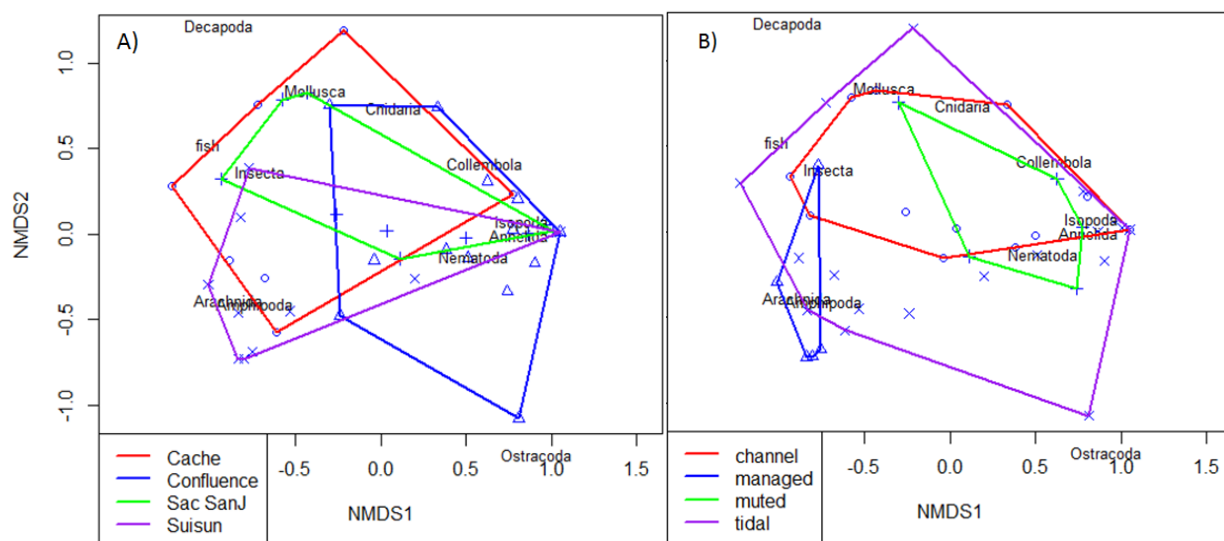


FIGURE 37. RELATIVE ABUNDANCE OF MAJOR TAXONOMIC GROUPS OF MACROINVERTEBRATES IN EACH HABITAT BY SITE AND REGION.



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FIGURE 38. NMDS ON BENTHIC DATA STRESS = 0.124 WITH A) HULLS AROUND REGION OF THE ESTUARY. CENTROIDS OF HULLS ARE SIGNIFICANTLY DIFFERENT ($R^2 = 0.29$, $p = 0.003$), AND B) HULLS AROUND SITE TYPE. CENTROIDS OF HULLS ARE SIGNIFICANTLY DIFFERENT ($R^2 = 0.20$, $p = 0.01$).

TABLE 30. BENTHIC SAMPLES PERMANOVA. 999 FREE PERMUTATIONS, TERMS ADDED SEQUENTIALLY. REGION OF THE ESTUARY HAD A HIGHLY SIGNIFICANT EFFECT ON COMMUNITY COMPOSITION, BUT SITE TYPE DID NOT.

Term	DF	Sums of sqs.	Mean sqs.	f- value	R ²	p-value	
Region	3	2.1296	0.7099	4.5805	0.24124	0.001	*
Site Type	3	0.9863	0.3288	2.1215	0.11173	0.05	.
Site	3	0.9075	0.3025	1.9519	0.1028	0.072	
Residuals	31	4.8043	0.155	0.54422			
Total	40	8.8277	1				

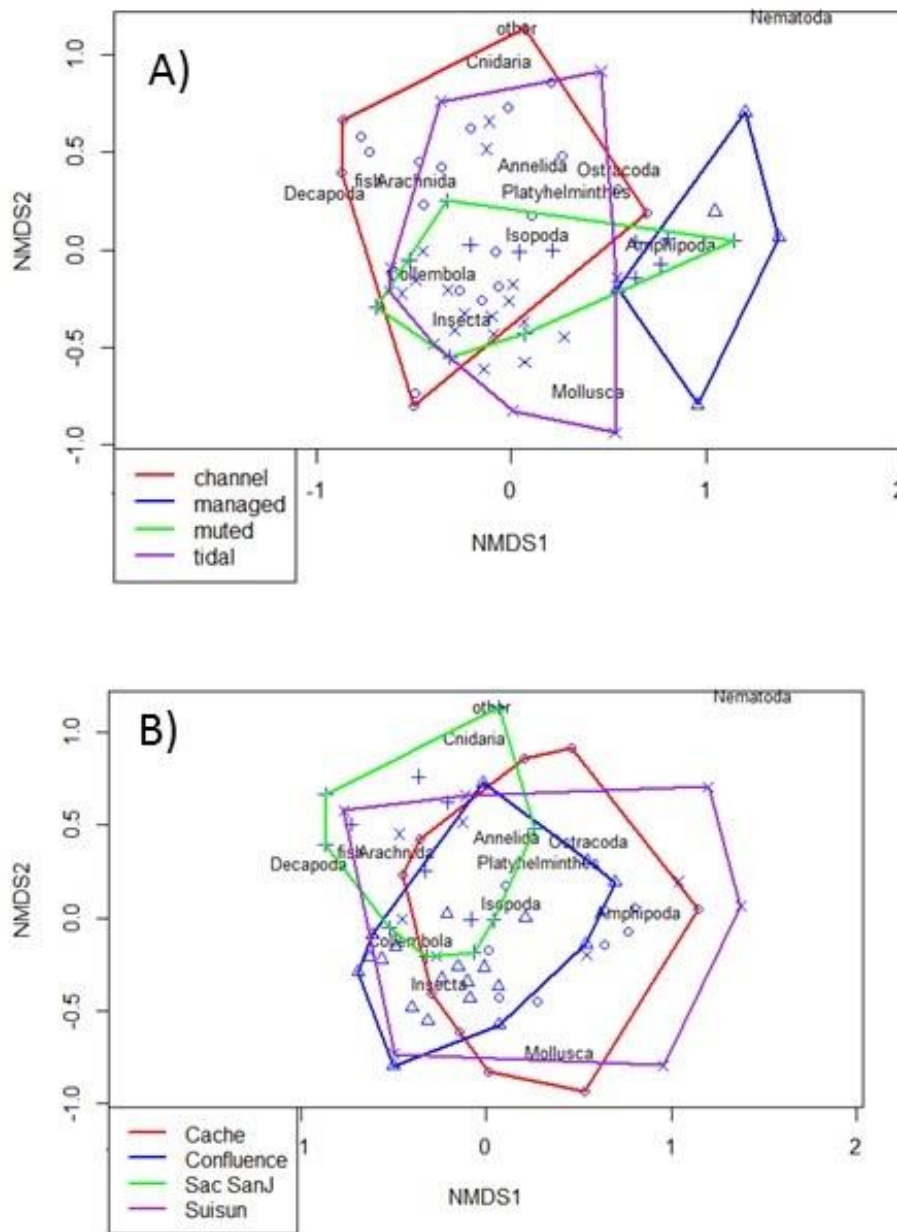


FIGURE 39 NMDS ON MYSID SAMPLES, STRESS = 0.196 WITH A) HULLS AROUND SITE TYPES. CENTROIDS ARE SIGNIFICANTLY DIFFERENT, $R^2 = 0.293$, $p = 0.001$. AND B) HULLS AROUND REGIONS OF THE ESTUARY. CENTROIDS OF HULLS ARE SIGNIFICANTLY DIFFERENT, $R^2 = 0.183$, $p = 0.001$.

TABLE 31. MYSID SAMPLES PERMANOVA. 999 FREE PERMUTATIONS, TERMS ADDED SEQUENTIALLY. SITE TYPE AND REGION OF THE ESTUARY HAD A HIGHLY SIGNIFICANT EFFECT ON COMMUNITY COMPOSITION. SITE (USED AS A BLOCKING TERM) ALSO HAD A HIGHLY SIGNIFICANT EFFECT ON COMMUNITY COMPOSITION.

Term	DF	Sums of Sqs.	Mean Sqs.	f-value	R^2	p-value	
Region	3	1.541	0.514	4.036	0.133	0.001	*

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Site Type	3	2.261	0.754	5.924	0.195	0.001	*
Site	6	1.425	0.237	1.866	0.123	0.007	*
Residuals	50	6.362	0.127	0.549			
Total	62	11.588	1				

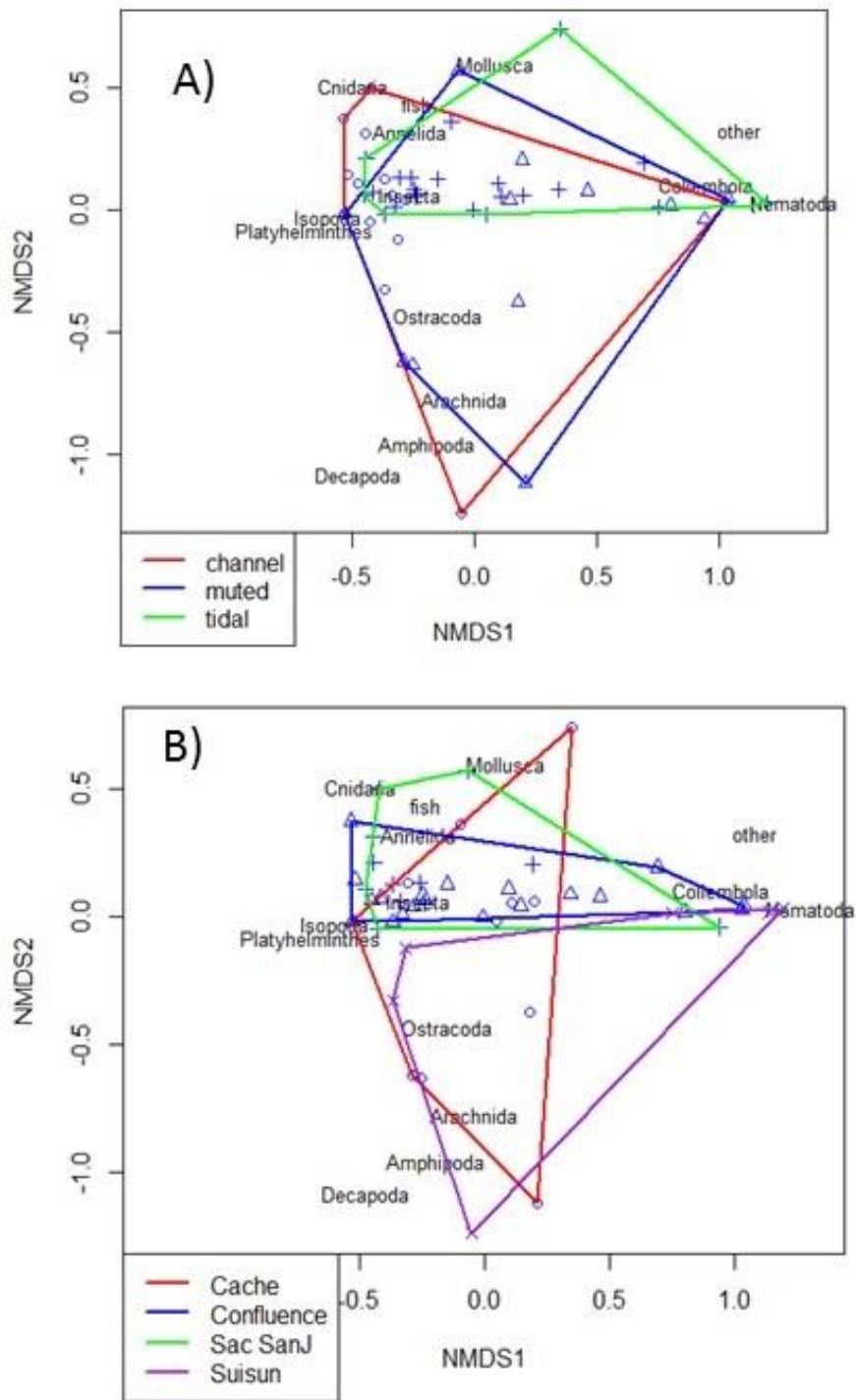


FIGURE 40. NMDS PLOT OF NEUSTON SAMPLE, STRESS = 0.107 WITH A) HULLS AROUND SITE TYPE. CENTROIDS OF HULLS WERE SIGNIFICANTLY DIFFERENT ($R^2 = 0.140$, $p = 0.012$) NOTE: WE DID NOT SAMPLE NEUSTON AT TULE RED DUE

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TO LOGISTICAL DIFFICULTIES, SO NO MANAGED WETLAND DATA IS SHOWN. AND B) HULLS AROUND REGION OF THE ESTUARY. CENTROIDS OF HULLS WERE SIGNIFICANTLY DIFFERENT ($R^2 = 0.153$, $p = 0.018$).

TABLE 32. NEUSTON PERMANOVA. 999 FREE PERMUTATIONS, TERMS ADDED SEQUENTIALLY. SITE TYPE AND REGION OF THE ESTUARY HAD A HIGHLY SIGNIFICANT EFFECT ON COMMUNITY COMPOSITION. SITE (USED AS A BLOCKING TERM) ALSO HAD A HIGHLY SIGNIFICANT EFFECT ON COMMUNITY COMPOSITION.

Term	DF	Sums of Sqs.	Mean Sqs.	f-value	R ²	p-value	
Region	3	0.946	0.315	5.003	0.168	0.001	*
Site Type	2	1.005	0.503	7.974	0.178	0.001	*
Site	5	1.235	0.247	3.918	0.219	0.001	*
Residuals	39	2.458	0.063	0.436			
Total	49	5.644	1				

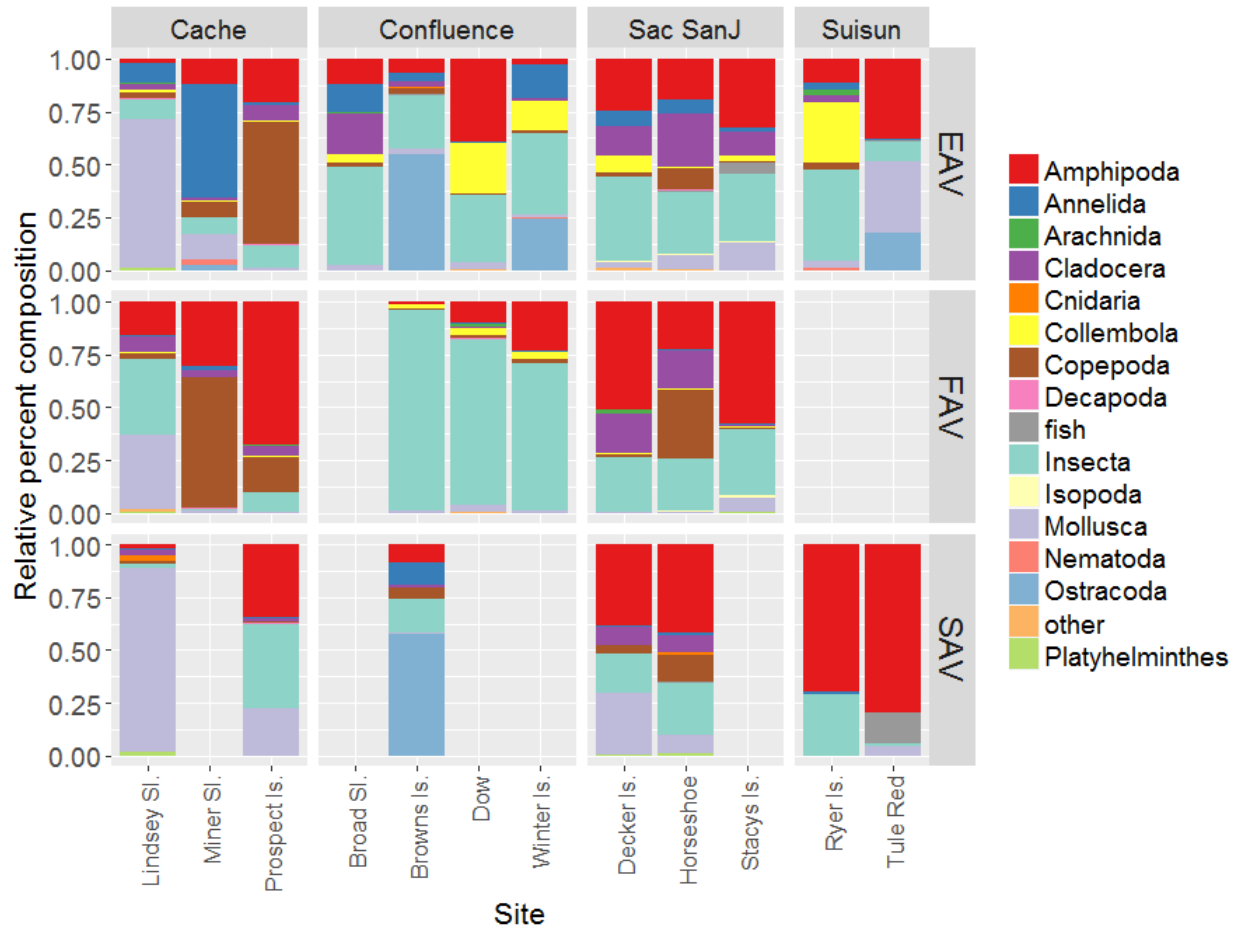
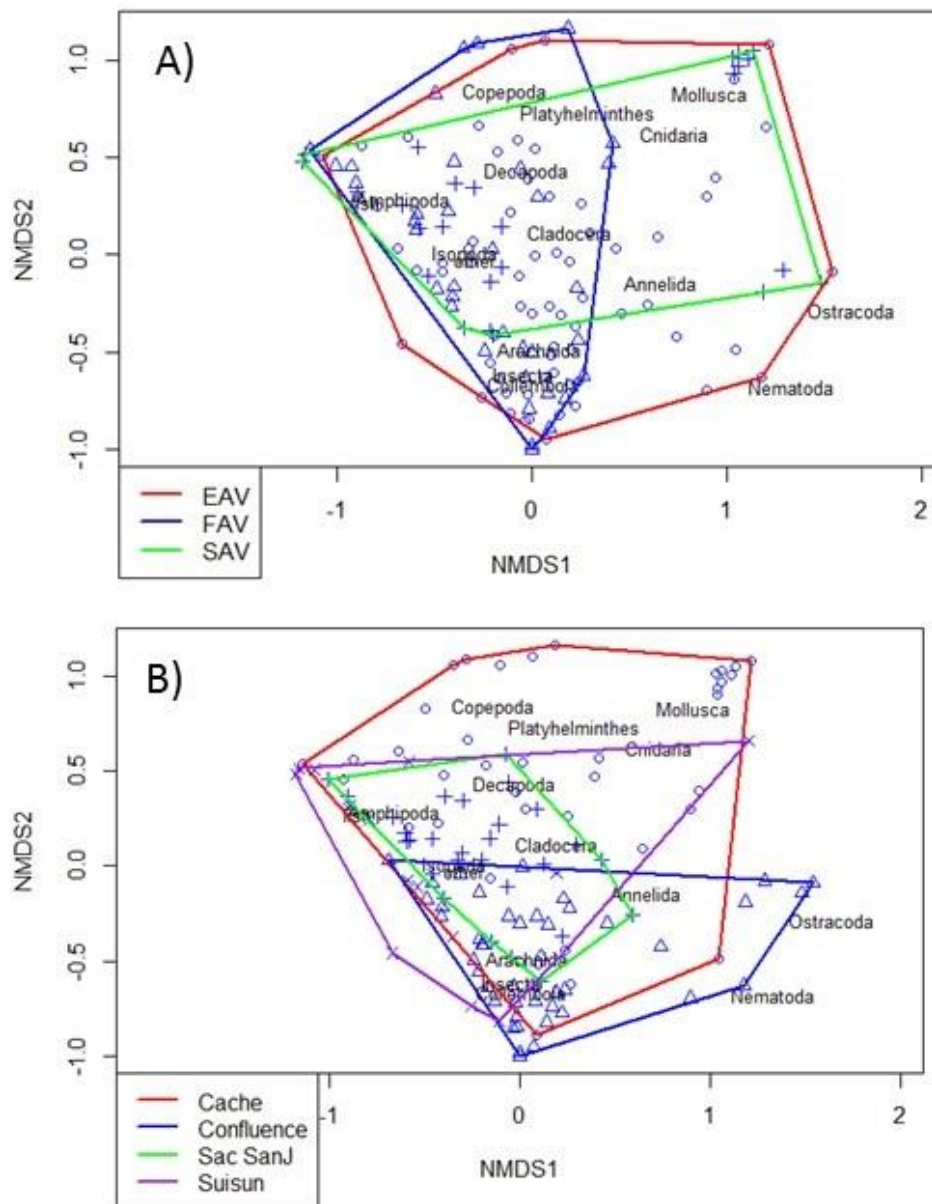


FIGURE 41. RELATIVE PERCENT COMMUNITY COMPOSITION OF MAJOR MACROINVERTEBRATE AND ZOOPLANKTON TAXA IN SWEEP NET SAMPLES. NOT ALL VEGETATION TYPES WERE PRESENT AT ALL SITES, CAUSING AN UNBALANCED DESIGN.



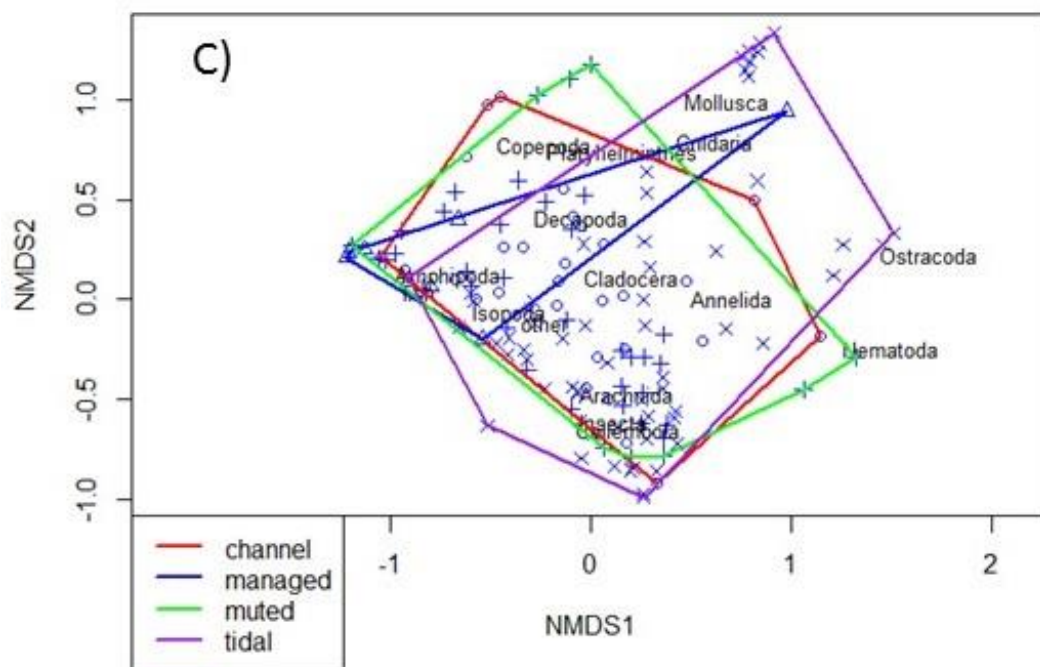


FIGURE 42. SWEEP NET NMDS. STRESS = 0.193. A) HULLS AROUND VEGETATION TYPES. CENTROIDS OF HULLS WERE SIGNIFICANTLY DIFFERENT ($R^2 = 0.079$, $p = 0.001$) B) HULLS AROUND REGIONS. CENTROIDS OF HULLS WERE SIGNIFICANTLY DIFFERENT ($R^2 = 0.300$, $p = 0.001$) C) HULLS AROUND SITE TYPES. CENTROIDS OF HULLS WERE SIGNIFICANTLY DIFFERENT ($R^2 = 0.103$, $p = 0.001$).

TABLE 33 SWEEP NET PERMANOVA 999 FREE PERMUTATIONS, TERMS ADDED SEQUENTIALLY. VEGETATION TYPE, SITE TYPE, AND REGION OF THE ESTUARY ALL HAD A HIGHLY SIGNIFICANT EFFECT ON COMMUNITY COMPOSITION. SITE (USED AS A BLOCKING TERM) ALSO HAD A HIGHLY SIGNIFICANT EFFECT ON COMMUNITY COMPOSITION.

Term	DF	Sums of Sqs.	Mean Sqs.	f-value	R ²	p-value	
Vegetation Type	2	2.140	1.070	8.620	0.073	0.001	*
Site Type	3	3.351	1.117	8.997	0.115	0.001	*
Region	3	4.986	1.662	13.387	0.171	0.001	*
Site	5	3.317	0.663	5.344	0.114	0.001	*
Residuals	124	15.394	0.124	0.5274			
Total	137	29.187	1				

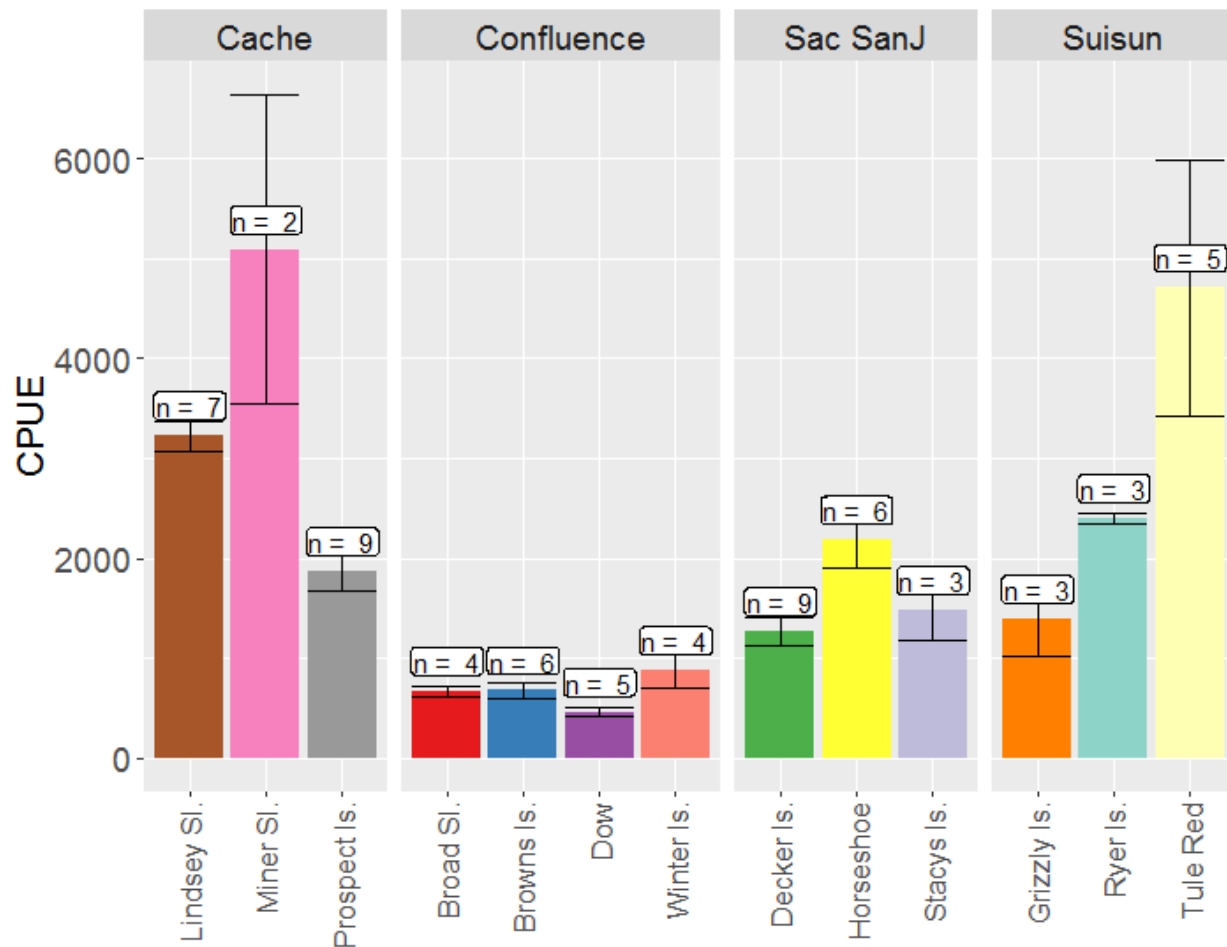


FIGURE 43. MEAN TOTAL CPUE (+/- 1 SEM) OF ZOOPLANKTON COLLECTED DURING THE SPRING SAMPLING PERIOD AT EACH SITE IN DIFFERENT REGIONS OF THE ESTUARY.

TABLE 34. FIXED EFFECTS OF GLMM OF LONG-TRANSFORMED ZOOPLANKTON CPUE. CACHE SLOUGH HAD SIGNIFICANTLY HIGHER ZOOPLANKTON CATCH THAN THE CONFLUENCE OR LOWER SACRAMENTO. THERE WAS NO SIGNIFICANT EFFECT OF WETLAND TYPE.

Term	DF	Estimate	SE	t-value	p-value	
Intercept: Cache, Channel	5.02	7.972	0.319	25.014	<0.0001	
Region: Confluence	3.73	-1.438	0.298	-4.832	<0.0001	*
Region: Sac San J	2.95	-0.625	0.300	-2.085	0.041	*
Region: Suisun	7.06	-0.662	0.436	-1.518	0.134	
SiteType: managed	6.45	0.236	0.552	0.428	0.670	
SiteType: muted	3.99	-0.600	0.310	-1.935	0.058	.

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SiteType: tidal	6.20	-0.088	0.301	-0.293	0.770
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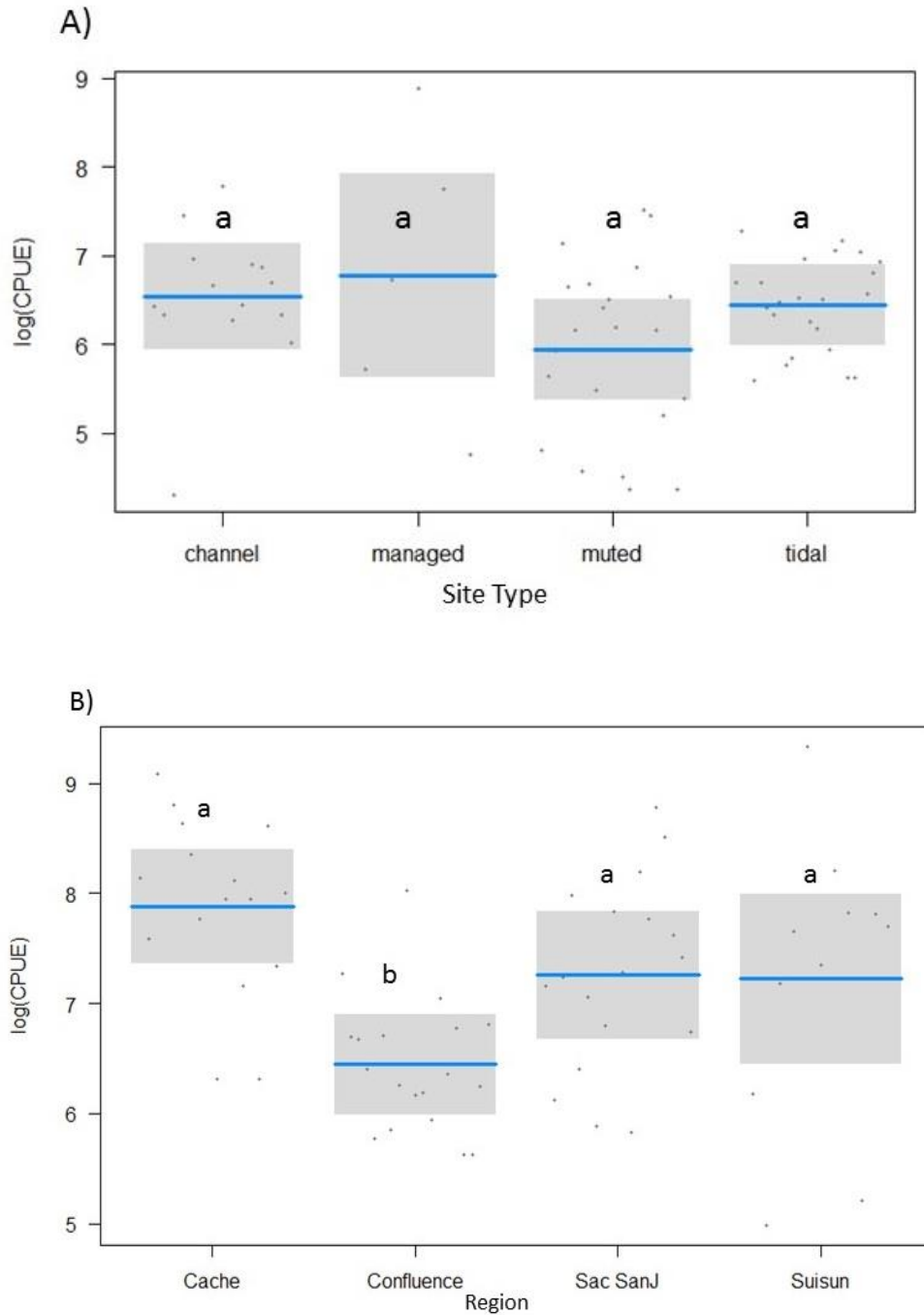


FIGURE 44. PARTIAL RESIDUAL PLOTS FOR GLM OF LOG ZOOPLANKTON CPUE VERSUS PREDICTOR VARIABLES. LETTERS INDICATE GROUPS THAT ARE NOT STATISTICALLY DIFFERENT ($p > 0.05$).

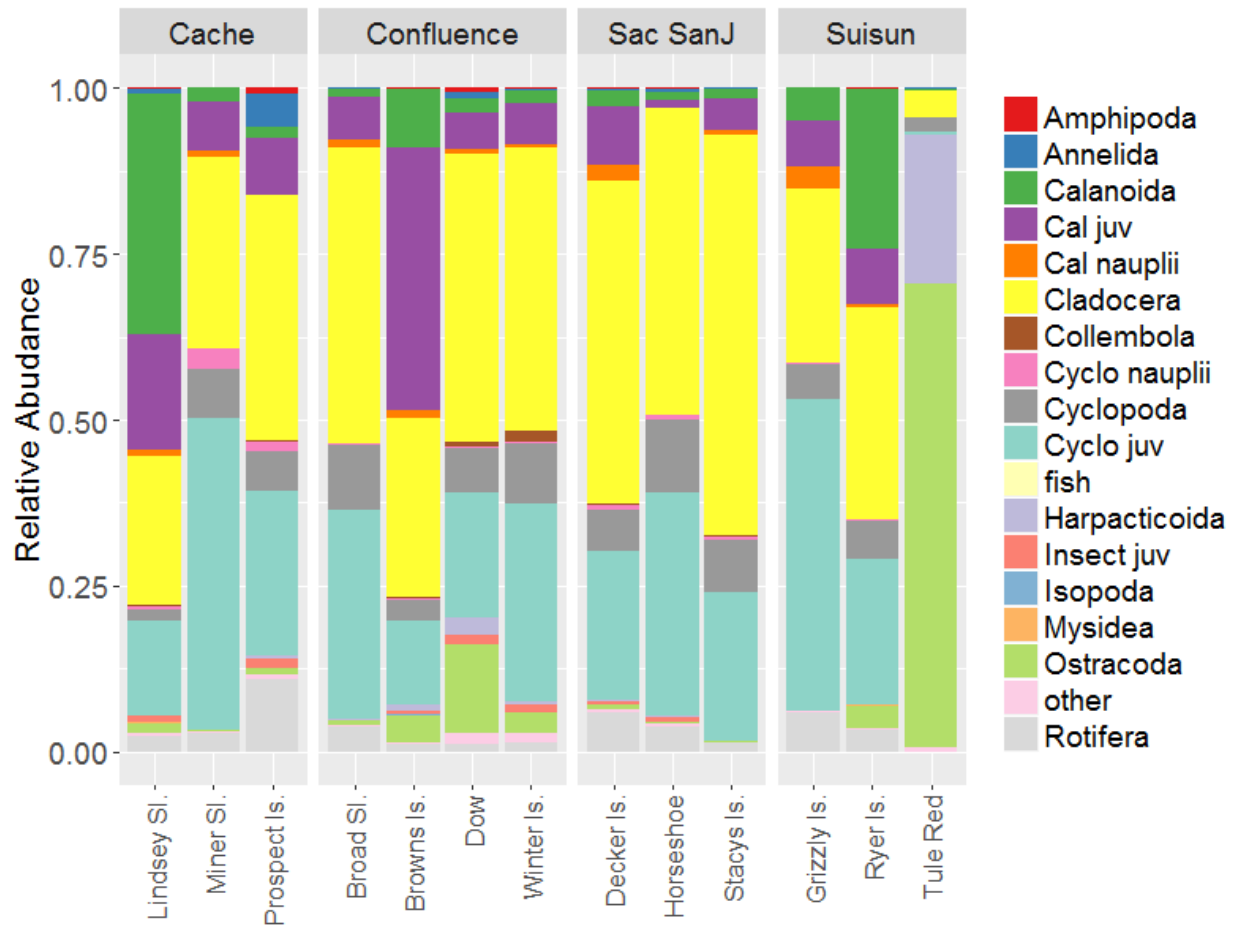


FIGURE 45. RELATIVE ABUNDANCE OF MAJOR GROUPS OF ZOOPLANKTON BY SITE AND REGION OF THE ESTUARY.

TABLE 35. PERMANOVA ON RELATIVE ABUNDANCE OF MAJOR ZOOPLANKTON TAXA IN SPRING SAMPLES. 999 FREE PERMUTATIONS, TERMS ADDED FIRST TO LAST. BOTH REGION OF THE ESTUARY AND SITE TYPE HAD A SIGNIFICANT EFFECT ON COMMUNITY COMPOSITION. SITE, ADDED AS A BLOCKING TERM, ALSO HAD A SIGNIFICANT EFFECT.

Term	DF	Sums of Sqs.	Mean Sqs.	f-value	R ²	p-value	
Region	3	1.3889	0.463	7.8171	0.18051	0.001	*
Site Type	3	2.1901	0.73	12.3265	0.28464	0.001	*
Site	6	0.9764	0.1627	2.7476	0.12689	0.001	*
Residuals	53	3.1389	0.0592	0.40795			
Total	65	7.6942	1				

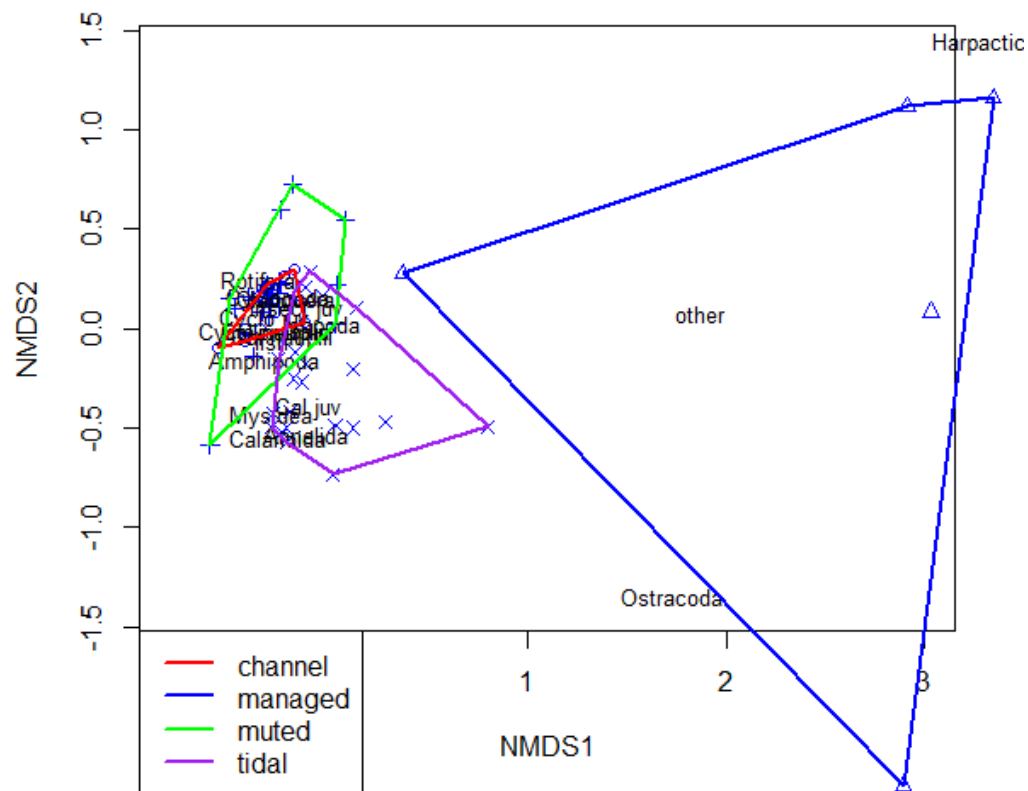


FIGURE 46. NMDS PLOT OF ALL ZOOPLANKTON SAMPLES COLLECTED DURING MARCH/APRIL OF 2017 (STRESS= 0.102), WITH HULLS AROUND SITE TYPES. THE “MANAGED” HULL CONSISTS OF SAMPLES FROM A SINGLE SITE (TULE RED), WHICH

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WAS DOMINATED BY OSTRACODS, HARPACTICOID COPEPODS, AND “OTHER” (SNAILS, NEMATODES, AND APHIDS), TAXA ABUNDANT AT OTHER SITES.

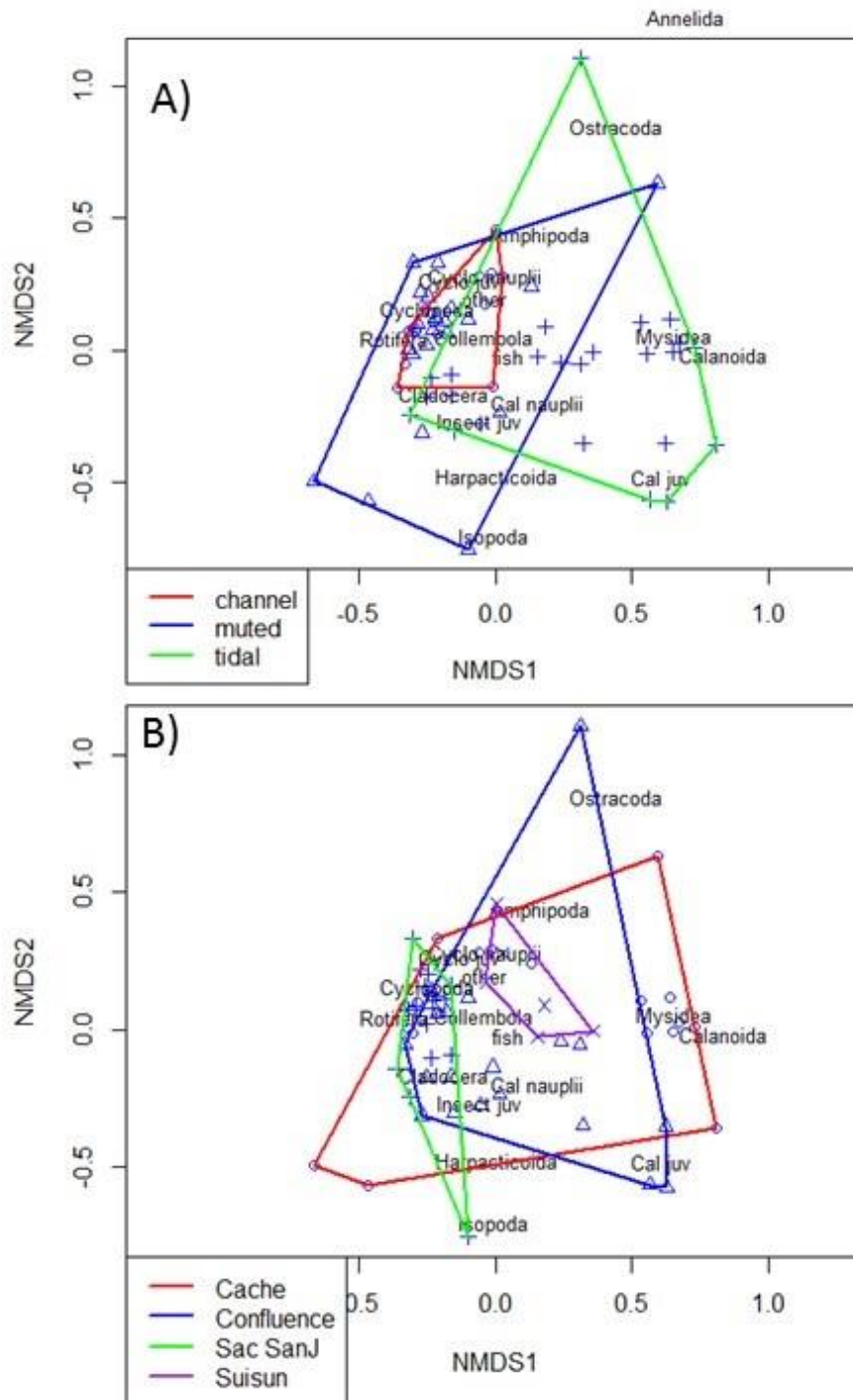


FIGURE 47. NMDS OF ZOOPLANKTON SAMPLES WITHOUT SAMPLES FROM TULE RED. STRESS = 0.1233. A) HULLS AROUND SITE TYPES, CENTROIDS WERE SIGNIFICANTLY DIFFERENT ($R^2 = 0.289$, $p = 0.001$). B) HULLS AROUND REGION OF THE ESTUARY, CENTROIDS WERE SIGNIFICANTLY DIFFERENT ($R^2 = 0.158$, $p = 0.008$).

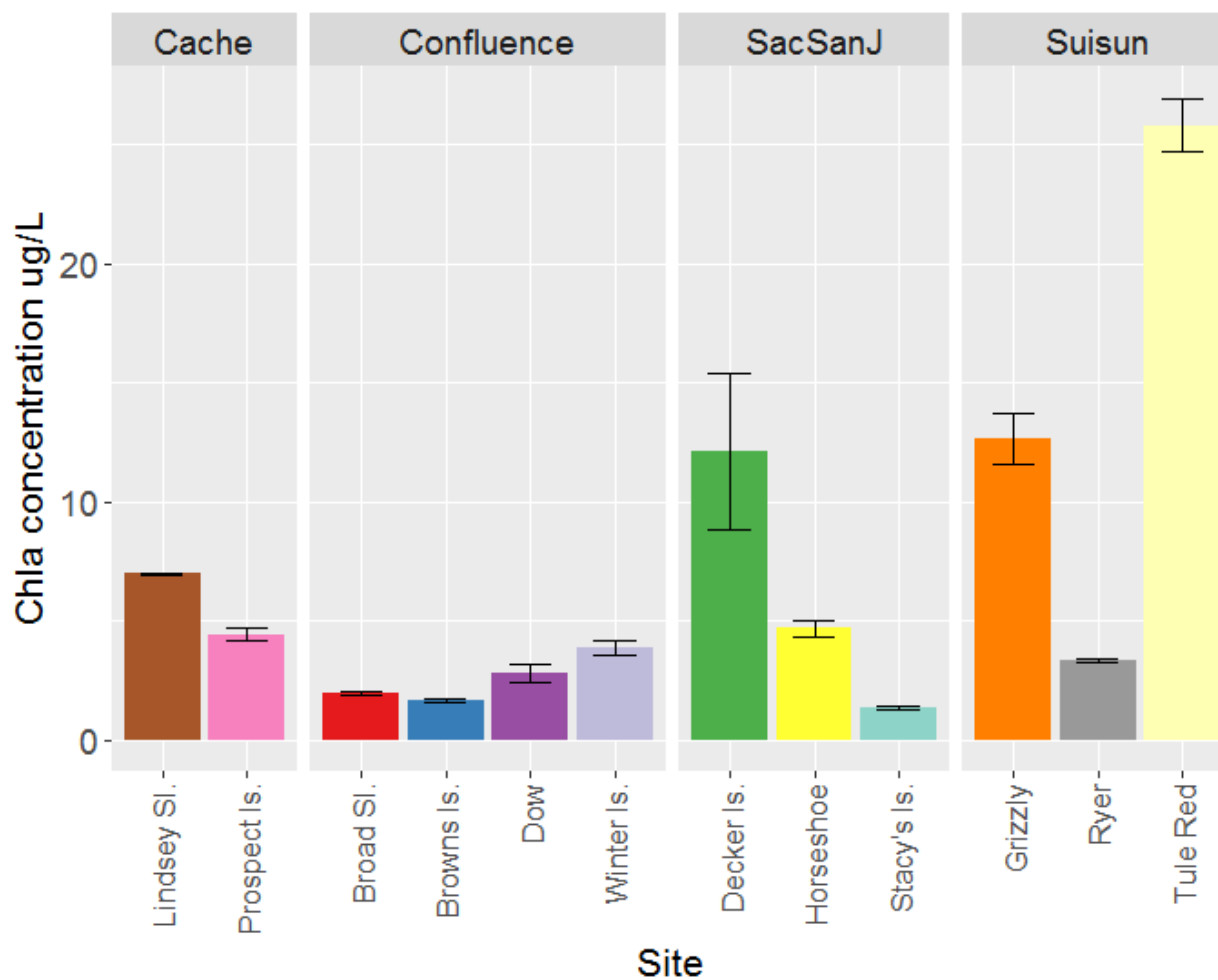


FIGURE 48. MEAN CHLOROPHYLL-A CONCENTRATIONS (+/- 1 SEM) FROM GRAB SAMPLES COLLECTED DURING ZOOPLANKTON AND MACROINVERTEBRATE SAMPLING.

TABLE 36. FIXED EFFECTS OF GLMM OF CHLOROPHYLL-A CONCENTRATIONS BETWEEN SITE TYPES AND REGIONS OF THE ESTUARY. SITE WAS USED AS A RANDOM EFFECT. TULE RED (MANAGED WETLAND) HAD SIGNIFICANTLY HIGHER CHLOROPHYLL THAN THE OTHER SITES, BUT THERE WERE NO OTHER SIGNIFICANT DIFFERENCES.

Term	Estimate	SE	DF	t-value	p-value
Intercept: Cache, Channel	6.20523	3.87251	4.4504	1.602	0.1771
Region: Confluence	-2.80255	3.31885	4.0898	-0.844	0.445

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Region: SacSanJ	0.09937	3.59484	4.5144	0.028	0.9791	
Region: Suisun	2.70709	4.23035	4.8033	0.64	0.5515	
Sitetype: Managed	16.87768	4.97297	4.8771	3.394	0.0201	*
Sitetype: Muted	1.29642	3.43329	4.6723	0.378	0.7223	
Sitetype: Tidal	-2.2673	2.95972	4.9848	-0.766	0.4783	

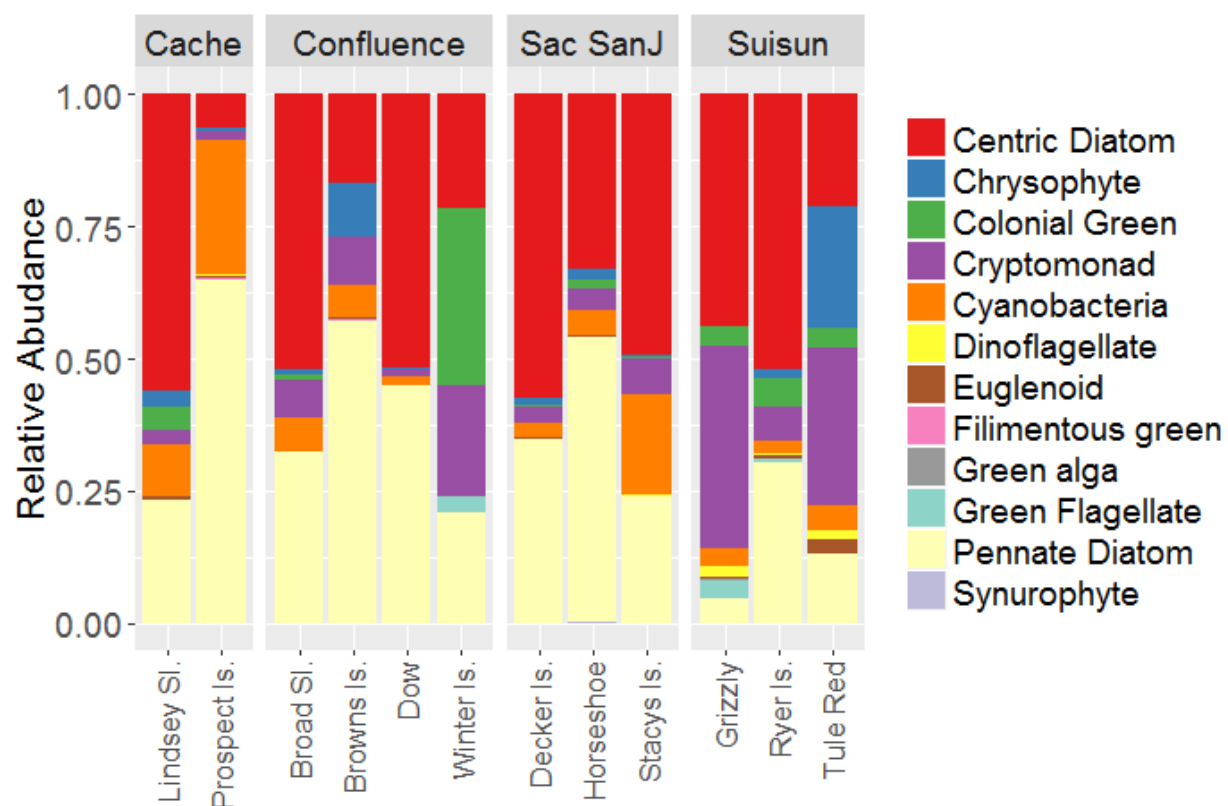


FIGURE 49. RELATIVE ABUNDANCE OF MAJOR PHYTOPLANKTON GROUPS, BY REGION OF THE ESTUARY AND SITE.

TABLE 37. PERMANOVA ON RELATIVE ABUNDANCE OF MAJOR PHYTOPLANKTON TAXA IN SPRING SAMPLES. 999 FREE PERMUTATIONS, TERMS ADDED FIRST TO LAST. BOTH REGION OF THE ESTUARY AND SITE TYPE HAD A SIGNIFICANT EFFECT ON COMMUNITY COMPOSITION. SITE, ADDED AS A BLOCKING TERM, ALSO HAD A SIGNIFICANT EFFECT.

Term	DF	Sums of Sqs.	Mean Sqs.	f-value	R ²	p-value	
Region	3	1.616	0.539	6.601	0.234	0.001	*
Site Type	3	1.048	0.349	4.281	0.152	0.001	*
Site	5	1.316	0.263	3.224	0.190	0.001	*

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Residuals	36	2.938	0.082	0.425
Total	47	6.918	1	

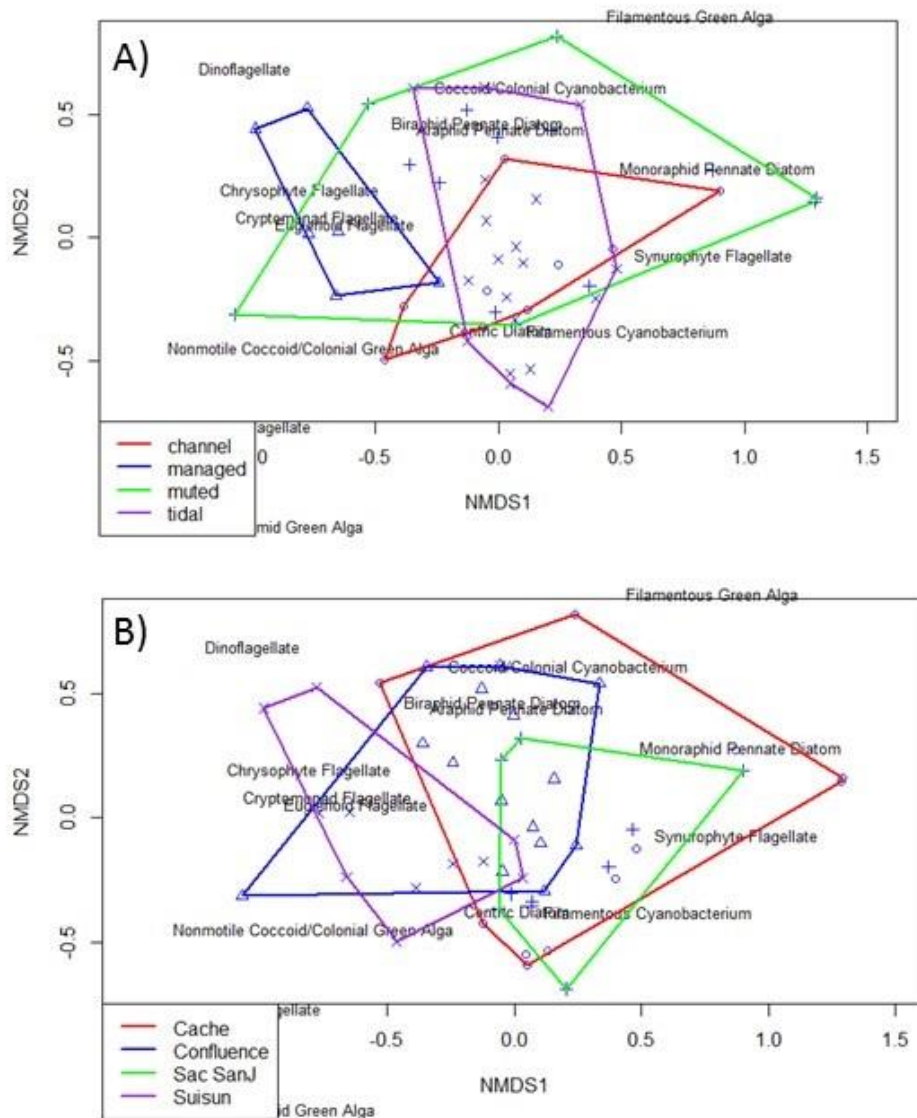


FIGURE 50. NMDS ON RELATIVE ABUNDANCE OF MAJOR GROUPS OF PHYTOPLANKTON (STRESS = 0.193) WITH A) HULLS AROUND SITE TYPE. CENTROIDS OF HULLS ARE SIGNIFICANTLY DIFFERENT ($R^2 = 0.234$, $p = 0.001$). AND B) WITH HULLS AROUND REGION OF THE ESTUARY. CENTROIDS OF HULLS ARE SIGNIFICANTLY DIFFERENT ($R^2 = 0.282$, $p = 0.001$).

TABLE 38. THE COEFFICIENT OF VARIATION (CV) IN CATCH PER UNIT EFFORT (CPUE) WITHIN SITES AND ACROSS SITES BY HABITAT TYPE. IF THE WITHIN-SITE CV IS HIGHER THAN THE CV OF THE SITE MEANS (IN RED), IT WILL BE DIFFICULT TO

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MAKE INFERENCES ON DIFFERENCES BETWEEN SITES. IF THE ACROSS-SITE CV IS HIGHER THAN THE WITHIN-SITE CV (IN GREEN), IT IS MORE LIKELY THAT WE WILL BE ABLE TO DIFFERENTIATE BETWEEN SITES.

Habitat	Mean within-site CV	CV in mean CPUE across sites
benthic	0.83248	1.593372
mysids	1.004067	3.434725
neuston	0.834965	1.299659
vegetation (all)	1.256011	1.065015
EAV	1.071	1.664
SAV	1.086	0.927
FAV	0.788	0.756
zooplankton	0.6554	0.7402
chlorophyll- <i>a</i>	0.352	1.0391

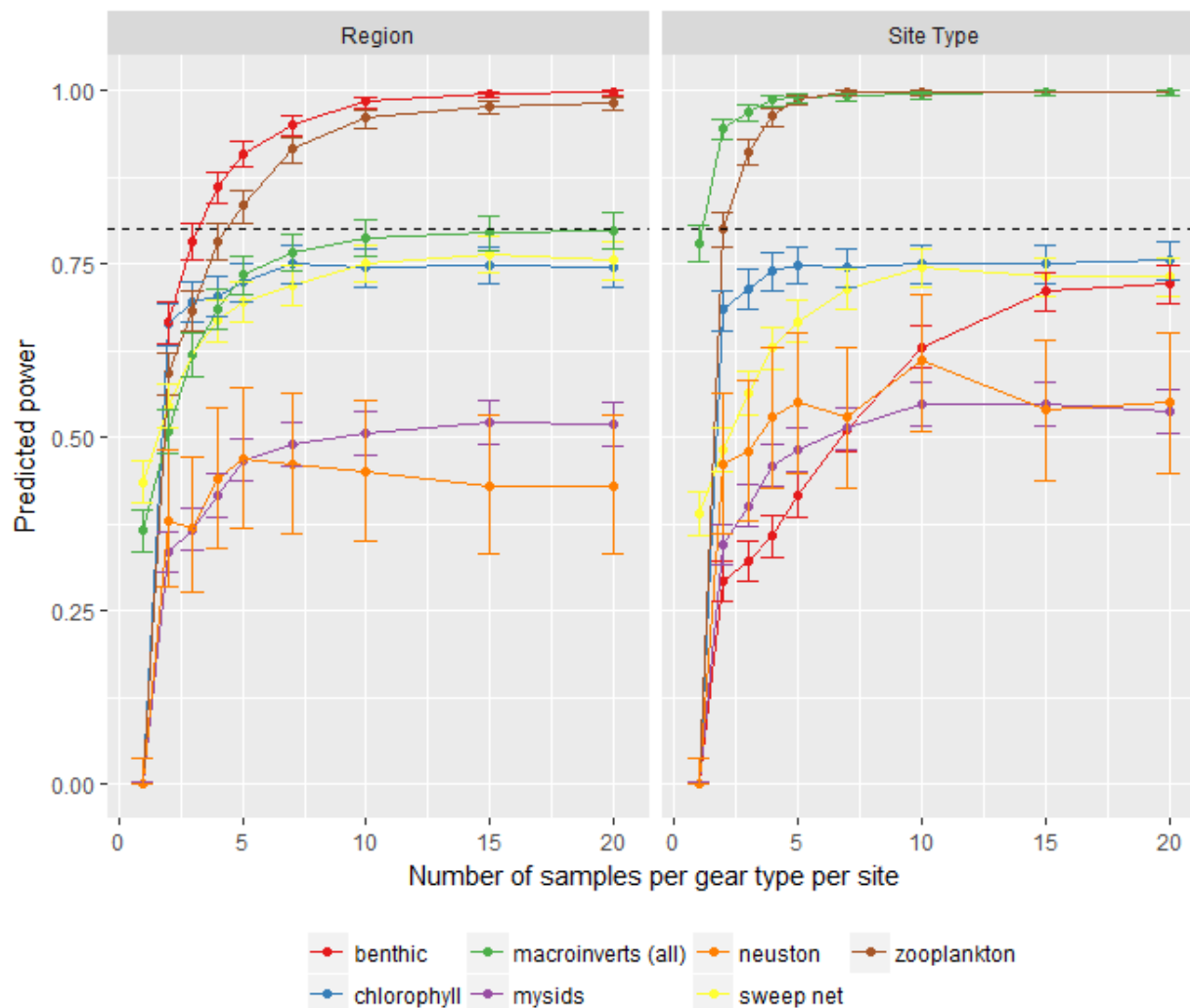


FIGURE 51. PREDICTED POWER (+/- 1 SEM) VERSUS SAMPLE SIZE FOR EACH SAMPLE TYPE NECESSARY TO DETECT A LN(1) DIFFERENCE IN CPUE AT THE $\alpha = 0.05$ LEVEL. RESULT OF 999 SIMULATIONS FOR EACH SAMPLE SIZE.

TABLE 39. SAMPLES PER GEAR TYPE PER SITE NECESSARY TO DIFFERENTIATE BETWEEN A LN(1) DIFFERENCE IN CPUE BETWEEN GROUPS WITH 80% POWER FOR EACH COMMUNITY BASED ON A POST-HOC POWER ANALYSIS. SAMPLES WITH "NA" WERE TOO HIGHLY VARIABLE TO EXTRAPOLATE SAMPLE SIZE USING PERMUTATIONAL METHODS.

Food web element	Between region	Between site type
all macroinverts	1	5
benthic	4	20
mysids	NA	NA
neuston	NA	NA
vegetation	NA	20+
zooplankton	4	2
chlorophyll- <i>a</i>	20+	20+

Discussion

Wetlands continue to be a highly variable and diverse habitat for many links in the food web that supports listed fishes. We found some differences in relative abundance of certain groups of invertebrates and phytoplankton between wetland types and regions of the estuary, but found it more difficult to differentiate between CPUE of these groups between wetland type or region. However, we now have a better understanding of how many samples are necessary to answer important questions regarding the relative production of different taxa on restoration sites, existing wetlands, and surrounding channel habitat.

Variation and sample size

In previous years, we found relatively low sample sizes ($n=3-6$ per habitat type and site) allowed us to make inferences about differences in community composition. However, we could not make any inferences about differences in abundance or CPUE (Contreras et al. 2017). This year, with increased sample size, we again found it easy to detect differences in *relative* abundance (community composition), but could not always detect differences in an index of *actual* abundance (CPUE).

While phytoplankton, zooplankton, and macroinvertebrates were all highly variable, macroinvertebrates exhibited the highest within-site coefficient of variation in CPUE, especially samples in vegetation (Table 38). This means that while vegetated habitat may be providing an important source of invertebrates, it may not be cost-effective to sample with enough replication to make inferences on differences between abundance of invertebrates in vegetation at restoration sites versus invertebrates on vegetation in channel habitat. The mysid samples also have very high within-site variation, making it difficult to make any inferences. However, when all of the macroinvertebrate data were combined, we could detect significant differences in CPUE between site types with only a few samples per gear type (Figure 51, 39). Therefore, while we will reduce the number of sweep-net samples taken in each vegetation type in the future, we will still collect some sweep-net samples in order to measure the contribution of vegetated habitat to overall wetland macroinvertebrate productivity. Much greater replication may be necessary to differentiate CPUE between regions of the estuary, but differences between site type are more important for addressing our restoration hypotheses (Figure 51, Table 39).

The mesozooplankton net (150 μm) had higher predicted power than the mysid net for detecting trends between site types and regions (Figure 51). This is supported by EMP's data, which also finds higher variability in mysid than in zooplankton samples (Hennessy et al. in prep). However, we did not detect differences between site types in the zooplankton data (Figure 44), and the observed differences were smaller than those used for the power calculations. This may indicate site types truly did not have a biologically significant difference in zooplankton CPUE. In the future, we may be able to reduce the number of zooplankton samples, particularly during the spring sampling bout. Because zooplankton, particularly this smaller size class, are transported mainly by the currents rather than under their own power, they may tend to cluster less on the landscape (Burks et al. 2002), and be more homogeneous between wetlands and the surrounding channel (see Part 1. Channel versus Shallow Water Comparisons). Bollens et al. (2014) found a similar result, with zooplankton samples from three channels in the same marsh forming similar communities.

Chlorophyll-*a* was the least variable indicator within sites, but had high variation between sites (Figure 48, Table 38). All the chlorophyll samples within a given site were collected on the same day, but because phytoplankton have such a high turnover rate, it may be more informative to take samples

spread out over a longer time period (as suggested by Kraus et al. (2017)). Instead of looking for spatial variation in chlorophyll within a site, future FRP monitoring will examine temporal variation through use of continuously recording sondes stationed at one or two locations within a site.

Data were somewhat unbalanced in 2017, so some of the difficulty in detecting differences may be alleviated by better sampling design and multiple years of data. Lessons learned from field work in 2017 allowed us to be much more efficient in 2018 and collect a relatively well-balanced data set. We will re-evaluate our questions on sample size and replication on a yearly basis, and scale back when necessary if we can achieve our goals with less effort. Despite difficulties in differentiating between CPUE of these groups, differences in community composition (discussed below) are easier to detect, and are still useful in informing relative food-web benefits of different site types.

Differences between regions

The four regions analyzed in this study are along the “North Delta Arc” of native fish habitat and diversity. While all regions may provide habitat, they have different hydrological and salinity regimes that translate to different food web characteristics and therefore different benefits to native fishes. We found major regional differences in zooplankton abundance, but not in chlorophyll or overall macroinvertebrate abundance. However, lack of a significant difference is more likely due to low sample size and the single sampling event rather than a lack of patterns. Other studies have found consistent trends of higher chlorophyll-*a* at freshwater sites, particularly in the Central and South Delta (Baxter et al. 2015; Jassby 2008; Kayfetz and Kimmerer 2017). The EMP mysid data finds higher abundances of mysids in the West Delta, Suisun Marsh, and the Confluence when compared to Suisun Bay (Hennessy et al. in prep). While overall abundance was similar between regions, there were major differences in community composition of all invertebrate communities between all these regions, which is supported by our previous years’ data and multiple studies by other researchers (Howe et al. 2014; Thompson et al. 2013).

Cache: The Cache Slough Complex, which includes Lindsey Slough, Prospect Island, and Miner Slough, is at the northeast edge of the Delta, and is strongly influenced by the Yolo Bypass and Sacramento River inputs (see Figure 30). This region had relatively more mollusks than other regions, particularly *Corbicula* (clams) in benthic samples and a variety of snails in vegetated samples. *Corbicula* is an invasive clam that has the potential to reduce phytoplankton availability, and has been found frequently in the area (Simenstad et al. 2013; Young et al. 2016). *Corbicula* may reduce phytoplankton standing stock locally (Lucas et al. 2002), but they have not had the sweeping food web effects that *Potamocorbula* has had downstream because *Corbicula*’s grazing rate is approximately four times lower than *Potamocorbula*’s grazing rate (Crauder et al. 2016).

Despite the abundance of clams, chlorophyll concentrations in Cache Slough were similar to other regions (Figure 48), and the majority of the phytoplankton were high-food-value diatoms (Figure 49). This is contrary to previous studies of phytoplankton in the region that found green algae and chrysophytes to outnumber diatoms in the nearby wetlands of Liberty Island (Lehman et al. 2010). While there were many diatoms, there were also more cyanobacteria in the phytoplankton of Cache Slough than in other areas. Cyanobacteria are considered poor food resources for zooplankton, and have the potential to cause toxicity (Galloway and Winder 2015; Ger et al. 2010). Cyanobacteria blooms, particularly *Microcystis*, are more frequent later in the season as the water warms (Lehman et al. 2013),

and are more common in drought years (Lehman et al. 2017), so may impact other regions later in the year or in future years.

Cache Slough zooplankton samples did have significantly higher CPUE than the confluence, and higher (though not significantly higher) CPUE than Sacramento-San Joaquin or Suisun (Figure 44), in keeping with previous research indicating higher zooplankton abundance in freshwater reaches of the Delta (Hammock et al. 2017; Kimmerer et al. 2018). There were also differences in community composition, with more adult calanoid copepods, annelids, and rotifers in the Cache region. Adult calanoid copepods, in particular, are one of the most important food sources for Delta Smelt (Slater and Baxter 2014), and the high abundance of copepods in the Cache Slough Complex may be supporting the resident population of Delta Smelt observed in this area (Sommer and Mejia 2013).

Sacramento-San Joaquin: The next region on the ecocline from the headwaters to the Bay was the Sacramento-San Joaquin River region (Figure 30). Decker Island, on the Sacramento River, has been characterized as a “hot spot” for Delta Smelt (Sommer and Mejia 2013). However, zooplankton samples had fewer calanoid copepods that Delta Smelt like to eat than in other regions (Figure 45). The high invertebrate production we found may be coming chiefly from the large bed of *Egeria*, rather than from tidal channels or open-water. Macroinvertebrate samples had a higher relative abundance of amphipods in the sweep-net samples and more insects in neuston samples (Figure 37, Figure 41), which may be due to the submerged vegetation. Amphipods and insects do occur in Delta Smelt diets in areas with high amounts of vegetation (Whitley and Bollens 2014), and make up a larger proportion of smelt diets now than historically (Baxter et al. 2015). However, vegetation can also provide refuge for non-native predators (Conrad et al. 2016). The current *Effects of Aquatic Macrophyte Control on Delta Smelt Habitat Study* is looking more closely at the interaction between aquatic weeds and the rest of the food web at Decker Island (J.L. Conrad, DWR, pers. comm.). Results of that study may be able to inform recommendations for managing weeds for optimal wetland productivity in future restoration sites.

This region was also a location for high “good” productivity, with chlorophyll-*a* concentrations above 10 µg/L in some samples at Decker Island (Figure 48), and a higher relative abundance of diatoms than any other region (Figure 49). However, while zooplankton had higher CPUE than the Confluence, it was lower than Cache Slough and had a lower percentage of calanoid copepods that generally flourish on diatoms (Kayfetz and Kimmerer 2017; Orsi 1995). The high percentage of Cladocera is to be expected, since Cladocera abundance is often positively correlated with chlorophyll-*a* (Muller-Solger et al. 2002), whereas the abundance of the most common copepod, *Pseudodiaptomus forbesi* is not (Kayfetz and Kimmerer 2017).

Confluence: In the Confluence, CPUE of macroinvertebrate samples in vegetation, neuston samples, and benthic samples were particularly high, though macroinvertebrates were not significantly higher overall (Figure 35, Figure 36). These samples were dominated by insects, amphipods, collembolans, and ostracods (Figure 37). Insects and collembolans are important in salmonid diets (Goertler et al. 2018), and since the confluence restoration sites are located at the “pinch point” where all salmon must pass on their way to the ocean (del Rosario et al. 2013), this might be an important source of food for their seaward migration.

Zooplankton CPUE was lower in the Confluence than any other region (Figure 43, Figure 44). Zooplankton samples had a higher relative abundance of juvenile calanoids and ostracods, and a lower abundance of adult calanoids (Figure 45). This is similar to studies of zooplankton from Browns Island

that found high ostracod and calanoid copepod densities (Bollens et al. 2014). The lower overall CPUE suggests the Confluence may not be as beneficial as Cache Slough in providing food for adult smelt, but may be important for larval and juvenile smelt, which rely extensively on juvenile copepods (Nobriga 2002; Slater and Baxter 2014). Longfin Smelt larvae have been found in the channels of Browns Island, so may be situated to benefit from this resource (Grimaldo et al. 2017). The particular data set presented here is from March and April, whereas the peak in abundance for many calanoid copepods is not until May or June (Merz et al. 2016), so these sites may provide greater benefits later in the year.

The Confluence is on the edge of the distribution for the invasive clam *Potamocorbula amurensis*, and while we did not detect this species in our benthic samples from the site, they have a seasonal peak in the summer, so may not have been detected by our April sampling (Crauder et al. 2016). While chlorophyll concentration in the Confluence was not significantly lower, no site in the Confluence had an average chlorophyll concentration above 4 µg/L (Figure 48), which may have limited zooplankton production (Hammock et al. 2016). Much of the phytoplankton that did occur at these sites was colonial green algae and cryptomonads, rather than diatoms (Figure 49). *Potamocorbula* clams are found more frequently in channels than in marshes (F. Feyrer, BDSC presentation), but regional abundances of clams may be partially to blame for the low zooplankton biomass, as they can affect phytoplankton and zooplankton biomass and community composition (Brown et al. 2016).

Suisun: The shallow waters of Grizzly Bay have historically been important Delta Smelt habitat due to high turbidity and shallow shoals. However, invasive clams have reduced phytoplankton and zooplankton abundance (Kimmerer and Lougee 2015; Kimmerer and Thompson 2014), and this region is not always a part of the low salinity zone during the fall when Delta Smelt are rearing (Brown et al. 2014).

Samples from Suisun included a large percentage of amphipods in benthic samples, particularly *Americorophium*, which is the most common amphipod in Delta Smelt diets (Slater and Baxter 2014). There were also higher percentages of Cnidaria in mysid samples. We did not find significant differences in zooplankton abundance, but long-term monitoring has found relatively low zooplankton abundance in Suisun Bay (Hennessy et al. In prep). Delta Smelt in this region often have high stomach fullness and foraging efficiency (Hammock et al. 2017), so the relatively modest zooplankton production may be subsidized by the high benthic amphipod production we found in our samples.

Suisun also had high concentrations of chlorophyll-*a*, and while they were not statistically significantly higher than other regions, samples from both Grizzly Bay and Tule Red were above 10 µg/L (Figure 48). Phytoplankton in Grizzly Bay and Ryer Island had high relative abundance of centric diatoms (Figure 49), which are high-quality food for calanoid copepods (Kayfetz and Kimmerer 2017). Both the relatively high chlorophyll values and the high percentage of centric diatoms are in contrast to an earlier study that found low (< 4 µg/L) chlorophyll concentrations and very low abundance of centric diatoms (Kimmerer et al. 2012).

Differences between site types

The major goal of FRP is showing the effectiveness of tidal restoration in providing food web benefits to fishes. Therefore, we need to show differences between food web resources in different types of wetlands.

Channel Habitat: Overall, macroinvertebrate abundance was lower in channel habitat than wetland habitat (Figure 36). Community composition was also different; the mysid net picked up a higher percentage of larval fish and insects than other types of sites, neuston samples had a higher percentage of amphipods, and sweep-net samples had more insects and decapods than other types of sites (Figure 37). The high relative abundance of larval fish in channel samples with relatively low absolute abundance of macroinvertebrates means larval fish may have more trouble finding food in channels than when foraging in wetland habitat.

Zooplankton CPUE was not significantly different, however channel habitat did have a significantly higher percentage of cyclopoid copepods and fewer calanoid copepods when compared with muted tidal or tidal wetlands (Figure 44, Figure 45). Cyclopoids are generally considered less desirable for smelt food than calanoid copepods (Slater and Baxter 2014).

Tidal and muted tidal wetlands: Tidal and muted tidal sites were relatively similar, with no significant differences in macroinvertebrate or zooplankton CPUE, though some differences in community composition. Tidal wetland sites had a higher percentage of calanoid copepods than either channel or managed wetland habitat (Figure 45), though fully tidal sites had even higher calanoid abundance than muted tidal sites. Fully tidal sites also had higher relative abundance of mollusks (Figure 41), which may compete with zooplankton and amphipods for phytoplankton resources (Kimmerer and Thompson 2014).

There was no statistically significant difference in chlorophyll concentration between channel and tidal wetland sites (Figure 48), which was surprising given numerous other studies finding higher chlorophyll in wetlands (Montgomery et al. 2015; Muller-Solger et al. 2002).

Managed wetland: There was only one managed wetland in this year's dataset, so we cannot make conclusions about managed wetlands as a whole. However, Tule Red had significantly higher chlorophyll concentration and significantly higher macroinvertebrate CPUE (Figure 35, Figure 48).

Macroinvertebrate samples had extremely high abundances of ostracods and amphipods (Figure 37), dominated by large-sized (1-2 cm) *Eogammarus*, a taxon not seen at other sites in the region. Zooplankton community composition was vastly different from any other sites in our data set (Figure 46, Figure 45). Phytoplankton samples had high relative abundance of chrysophyte flagellates that were rarely seen anywhere else, and very few of the pennate diatoms that dominated other sites (Figure 49). The different community composition is most likely due to Tule Red's managed wetland hydrology and being hydrologically cut-off from the surrounding waterways. In future years, sampling in other managed wetlands will allow us to see whether the high abundance of these taxa are unique to Tule Red, or indicative of managed wetlands in general.

The extremely high densities of macroinvertebrates found in other studies of managed wetlands has prompted some researchers to suggest that restoration sites should be managed to prolong the hydroperiod and "cook" productivity (Hobbs et al. 2017; Williamson et al. 2015). While Tule Red had high primary productivity, little of it was the centric diatoms preferred by copepods (Kayfetz and Kimmerer 2017), and the zooplankton samples collected at Tule Red were dominated by ostracods and harpacticoid copepods (Figure 45), neither of which is considered preferred food for smelt (Slater and Baxter 2014). The higher relative abundance of calanoid copepods at tidal wetlands suggests that fully tidal sites have more potential to "cook" the right kind of food for smelt, as well as providing better access to the food by creating a site that is open continuously.

With this single, intensive sampling bout, we found many differences in the food webs of these sites. However, all of the communities we sampled will change seasonally and inter-annually. From our shallow/channel comparison study, we saw how abundance of zooplankton and mysid samples increase over the course of the spring (Figure 16), most likely to a summer peak. Benthic invertebrates, surface invertebrates, epiphytic invertebrates, and phytoplankton also change over the course of the year and between years. Therefore, the patterns we found in community composition and abundance between regions and site types should be considered preliminary, and only applicable to spring of 2017 (a wet year). We look forward to increasing our data set with multiple years of data to see which of these trends persist over the long-term.

Endangered Species Act Take

A total of 5 Delta Smelt and 1 Longfin Smelt were caught during this study by the lampara net (Table 40) outside Tule Red in Grizzly Bay. All catches were reported to IEP as soon as possible after collection. All specimens were transferred to Jim Hobbs at UC Davis for otolith analysis.

TABLE 40. ESA FISH CAUGHT IN 2017.

Year	Month	Location	Sample type	Species	Catch
2017	6	Grizzly Bay	Lampara	Delta Smelt	2
2017	7	Grizzly Bay	Lampara	Longfin Smelt	1
2017	9	Grizzly Bay	Lampara	Delta Smelt	2
2017	10	Grizzly Bay	Lampara	Delta Smelt	1

Data Storage and Availability.

Data is currently stored in an Access database on CDFW's Stockton server. Data can be exported as a flat-file and shared upon request. Direct data requests to Rosemary Hartman: Rosemary.Hartman@wildlife.ca.gov.

Acknowledgments

Funding for this study was provided by the State Water Project through the DWR Fish Restoration Program. The work was included in the 2017 Interagency Ecological Program Work Plan (Element number 2017-311). We would like to thank our fantastic Scientific Aides: Sunny Lee, Walter (Kyle) Griffiths, and Mathew Crane for field help and a herculean amount of invertebrate sorting and identification. Also, our Senior Laboratory Assistant, Ryan Kok, for processing every single one of the zooplankton samples analyzed in this report. We would like to thank Matthew Siepert and Jared Mauldin for piloting our boats through muddy, weedy waters, and fixing the engines when they got clogged with SAV. We would also like to thank the multitude of volunteers who came out to help with this project, even when it involved sampling in the middle of the night. We would like to thank Dan Ellis for assistance with analysis and manuscript review.

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